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STRUCTURE AND MORPHOGENESIS OF TWO NEW SPECIES OF *HEXAMASTIX* FROM LIZARDS

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INTRODUCTION

As far as can be ascertained no flagellates belonging to the genus *Hexamastix* (Alexeieff) have been hitherto reported from reptiles. During an extensive survey of the intestinal flagellate fauna of amphibians and reptiles, I found 2 species of this trichomonad genus in several species of lizards. The individual hosts which harbored either one or both species of *Hexamastix* were always infected with several other flagellate genera, but never with *Monocercomonas* (= *Eutrichomastix*), even though the members of the latter genus were found to be among the most frequent inhabitants of the large intestine of squamate reptiles .

It is probable that the absence of any records of *Hexamastix* from reptiles in the literature has been caused by mistaken identification of this genus as *Monocercomonas*.

Although Kirby (1930) and Ludwig (1946), in their descriptions of *Hexamastix* from termites and crane-fly larvae respectively, made some references to the division process, neither of them presented a complete picture of morphogenesis of the mastigont organelles. During the present investigation, I was able to follow the development of these organelles in one of the species through the study of living organisms with the aid of phase contrast and through examination of protargol-stained permanent preparations.

MATERIALS AND METHODS

175 squamate reptiles belonging to 33 genera and 42 species, were found to harbor flagellates in the large intestine. Of these, 9 lizards, which belonged to 7 genera and as many species, showed an infection with 1 or 2 species of *Hexamastix*. The lizards were collected in the western part of the United States, primarily in California, and in Mexico.

Not even the majority of individuals collected in the same locality at the same time harbored *Hexamastix*. For example out of 3 skinks taken in a single area, only 1 was infected with this flagellate genus.

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[1] I wish to express my appreciation to Dr. Wade Fox for his invaluable help in collecting and identifying the reptilian hosts. Credit is also due to Messrs. C. B. Perkins and Charles Shaw of the San Diego Zoological Gardens for their aid in obtaining intestinal and fecal specimens from domestic and foreign reptiles.

This investigation was supported by Grants-in-Aid from Sigma Xi Resa Research Fund and from the Teaching Staff Research Council of the University of Massachuetts.

All the hosts, subsequently found to harbor *Hexamastix*, were caught in the field, brought to the laboratory, and their intestinal contents examined for the presence of parasites.

A strain of one of the species of *Hexamastix* was maintained in culture on Balamuth and Sandza's (1944) yolk infusion to which sterile horse serum was added (1:20). The culture was kept at room temperature and transferred routinely every 14 days. Much of the material employed in the observations of living organisms and for permanent preparations used in the study of divisions was derived from this culture.

Bouin's and Hollande's fixatives were used for preparations to be treated according to modified Bodian's protargol method (see Honigberg, 1951) or stained with iron hematein (or hematoxylin). Schaudinn's or weak Flemming's fluids were also employed prior to staining with iron hematein (or hematoxylin).

All observations of living material were made with the aid of phase contrast.

Hexamastix kirbyi n. sp.

(Figs. 1–32)

This species is named in honor of my late teacher, Professor Harold Kirby. It was found in 5 species of iguanid lizards: *Sauromalus obesus* (2 out of 4); *Uma notata cowlesi* (1 out of 2); *Sceloporus magister* (1 out of 1); *Uta stansburiana* (1 out of 3); and *Phrynosoma cornutum* (1 out of 1); as well as in the skink, *Eumeces gilberti* (1 out of 3).

All the hosts harbored several flagellate genera besides *Hexamastix*. However, while in the former 3 host species *H. kirbyi* was the only species of this genus, in the latter 3 it was accompanied by another species (see below).

One strain of *H. kirbyi*, from *Sauromalus obesus*, was maintained in culture.

DESCRIPTIVE ACCOUNT

(All measurements in microns).

The body is typically ovoidal (figs. 1, 3, 4, 8–11), although some individuals, particularly the larger ones, tend to be nearly spheroidal (fig. 12).

Fifty flagellates selected at random were measured in 1 population of each of 3 different strains. The body averaged 8.5(5–17.5) by 7(3.5–15.5) in a population from *Sauromalus obesus;* 8(5–12) by 6.5(2.5–10.5) in one from *Phrynosoma cornutum;* and 5.5(2.5–8.5) by 4(1.5–6) in one from *Eumeces gilberti.* A comparison of the 3 sets of figures indicates that differences in body size of different strains do not constitute satisfactory criteria in taxonomy of flagellate parasites. Furthermore, the size ranges and averages may differ significantly among different populations of a single strain, depending upon the rate of division in the population and physical and chemical conditions prevailing in the environment (in the host or in the culture medium).

In fully developed organisms there are 5 anterior flagella of unequal length and a single recurrent flagellum (figs. 1, 4, 7–12). All the flagella originate in a large basal granule complex (fig. 1). Occasionally, 2 granules are seen in this complex; one of them, the ventral, gives rise to all the anterior flagella and the other, more dorsally located, to the recurrent flagellum (fig. 8).

The anterior flagella, which typically terminate in rods or knobs (figs. 2–4, 9–12) are united in a short column at their base (figs. 1–6, 8, 10). The recurrent flagellum, which equals the anterior flagella in diameter, is characterized by an often quite long terminal filament or acroneme (figs. 3, 7, 9–12). In living organisms, the acroneme was seen occasionally to constitute over one quarter of the total length of the flagellum.

The longest anterior and the trailing flagella measured in 25 random organisms averaged in length 18.5(12–25.5) and 17(13–22.5); 21(10.5–26) and 20(12–25); and 13(12–15.5) and 14(10.5–19) in the strains from *Sauromalus obesus*, *Phrynosoma cornutum*, and *Eumeces gilberti* respectively.

Counts of the anterior flagella were made in the same populations in which the body and the flagella were measured. In the population from *Sauromalus obesus*, 100 random specimens were examined; of these, 40 had five; 30, four; and 10, only three flagella. Twenty specimens were unsuitable for counts. In the population from *Phrynosoma cornutum*, out of 220 specimens, 63% showed five, 12% four, and 4%, three flagella. In 21% of the organisms the flagella could not be counted. Out of 171 specimens examined in the population from *Eumeces gilberti*, 51% possessed five flagella, 27%, four, and 5%, only three. In 17% no accurate counts could be made.

The inequality in length and the differences in number of the anterior flagella may be explained by their behavior in division.

In the process of division the 6 flagella are equally distributed between the 2 daughter individuals. One of the daughters receives 2 of the anterior flagella and the recurrent flagellum (which can be recognized by its acroneme), while the other gets the remaining 3 anterior flagella (figs. 20–23). In many organisms in which the daughter nuclei are well formed and the new axostyles achieve considerable dimensions, no new flagella can be seen (fig. 25). Others show a short new flagellum prior to the completion of nuclear division (fig. 26). In some specimens the new recurrent flagellum appears first (fig. 26), but often short stumps of new flagella, of about equal length, are seen in both daughter mastigonts (figs. 28, 29). As the development of the mastigont systems progresses, the new flagella increase in length (fig. 30), but no additional flagella can be found prior to the completion of cytoplasmic division.

Several organisms contained 2 fully developed sets of organelles, each set with 3 anterior flagella (fig. 31). None of these specimens showed the extranuclear spindle ("paradesmose"). There is essentially very little difference between the very advanced stages of division of organisms that are about to undergo cytokinesis and the young double individuals which are incapable of cytoplasmic division, except for the absence of the extranuclear spindle in the latter group, and frequently (but by no means always) incomplete development of the distal parts of axostyles in the former. It is thus difficult to decide whether the specimens I saw represented double individuals or advanced division stages of forms capable of cytokinesis. The fact that occasionally the extranuclear spindle failed to stain with protargol in some of the earlier stages (figs. 21, 29) might suggest that at least some of these organisms belonged in the latter category.

As mentioned previously in this account, protargol-stained preparations of samples of most populations contain a small number of organisms with 3 anterior fla-

gella (fig. 3) and a larger number of medium-sized individuals with 4 flagella (fig. 2) in addition to many specimens equipped with 5 such organelles. The relative number of organisms equipped with less than 5 anterior flagella is directly proportional to that of division figures in a population. In some individuals that have 4 or 5 flagella, one of them is much shorter than the others (fig. 10). Similar differences in the number and length of the flagella are found in random samples of populations of living organisms. These variations depend upon the fact that the full flagellar complement is restored in solitary individuals which result from cytokinesis. The relative numbers of forms with 3 and 4 anterior flagella and the relative lengths of these organelles indicate that the fourth flagellum appears soon after cytoplasmic division and that the fifth does not grow out until the fourth one has reached a considerable length. The above conclusions were confirmed by observations of living individuals in which, on several occasions, I was able to follow the development of the fourth and fifth anterior flagella.

While some question exists as to the identity of the organisms that contain 2 fully developed mastigont systems, each with 3 anterior flagella, but without the extranuclear spindle, there can be no doubt that, in view of the foregoing observations, similar specimens, the mastigonts of which contain 4 (fig. 32) or 5 such flagella are double individuals incapable of cytokinesis.

The parabasal apparatus, situated somewhat dorsally and to the right of the nucleus (figs. 2, 3, 5–7, 9–12), originates in the basal granule complex to the right of the flagella (figs. 2, 10–12). It consists of a discoid parabasal body and a fine dorsal filament. However, the parabasal filament stains only in relatively few instances (figs. 9, 25–27, 30). The smaller parabasal bodies vary in shape, but show almost no undulations (figs. 2, 3, 5, 7). The larger ones exhibit a rather typical lateral undulation, which often gives the disk a somewhat bilobed appearance (figs. 6, 9, 11, 13, 16). Occasionally the large parabasals are almost saddle-shaped (fig. 12). The rim of the parabasal disk usually appears as a dark filament (figs. 3, 6, 9–17), while the central part of the body often takes a much lighter stain. Thus in some instances the parabasal could be incorrectly interpreted as being hollow (figs. 3, 5, 6, 11, 12, 14, 29); however, in many specimens the central part of the disk stains more heavily (figs. 9, 23, 25, 28). In certain views the parabasal body appears as a rod (fig. 15). The size of the parabasal body varies among and within different populations. As a rule, it is largest in the largest organisms (figs. 6, 12). This size relationship obtains also in division stages where the parabasals in giant dividing organisms (figs. 27, 29) are proportionately much larger than those found in normal specimens (figs. 28, 30). However, individuals in one strain (figs. 9, 11) may show larger parabasal bodies than organisms of similar dimensions in another strain (figs. 5, 7). The parabasal apparatus, not seen in preparations stained with iron hematein (figs. 1, 8), was observed not only in protargol-treated material, but also in many living organisms.

A large part of the old parabasal is discarded very early in division (fig. 18). Before the discarded part degenerates completely, a new small disk appears near the retained part of the parental parabasal (fig. 19). Since the material stained with protargol does not show the basal granule complex with clarity, it is impossible to ascertain whether in this stage the granule is divided. In any event, in slightly more advanced stages each of the small parabasals is associated with one of the

sister basal granules (fig. 20). From then on the parabasal bodies merely increase in size (figs. 22–31). They apparently continue to grow after completion of cytoplasmic division.

The hyaline axostyle originates in the basal granule complex (figs. 1–3, 8–10). The anterior part of the axostyle is expanded into a spatulate capitulum that is closely applied to the left ventro-lateral surface of the nucleus (figs. 4, 6, 7), and in some organisms extends for a considerable distance posterior to it (fig. 12). The trunk of the axostyle, a very slender rod (about 0.2 in diameter), runs through the approximate center of the body and projects from its posterior end for a considerable distance. The posterior projection, measured in 25 random specimens, averaged in length 5.5(2.5–8.5) in a population from *Sauromalus obesus,* 6.5(2.5–11) in one from *Phrynosoma cornutum,* and 4(2.5–5.5) in one from *Eumeces gilberti.* While in some fixed preparations the entire projecting part appears to taper uniformly (figs. 1, 3, 8), a study of other fixed specimens (figs. 9, 12) and of all living organisms indicates that only the distal part of the projection tapers gradually or more abruptly to a point.

The pelta is a crescent-shaped membrane situated near the anterior surface of the body. In some views it is relatively slender (figs. 3, 10, 11), in others quite robust (figs. 4, 7, 9). The membrane, delicately stained in many specimens (figs. 11, 12), stains very darkly in others (figs. 4, 7). It curves from the right-dorsal towards the left-ventral side of the body, ending near the antero-ventral surface of the nucleus (figs. 2, 5, 11) or somewhat anterior to it (fig. 9). The pelta remains in a close spatial relationship with the anterior part of the capitulum of the axostyle (figs. 2, 3, 4, 7, 9–12) and probably constitutes its extension. Many preparations stained with iron hematein (or hematoxylin) show the pelta quite clearly (figs. 1, 8). This structure can also be demonstrated in living organisms.

In very early stages of division the pelta shows signs of degenerative hypertrophy (fig. 19). Soon the capitulum of the axostyle, together with the pelta, becomes displaced in relation to the basal granules (fig. 20). Next both the pelta and the capitulum disappear and the degeneration of the parental axostyle proceeds toward the posterior end of the body (figs. 21–23). The degeneration is completed prior to the separation of the parental nucleus (figs. 24, 26).

Small primordia of the new axostyles appear in organisms in which a large remnant of the parental axostyle is still present (fig. 22). Each primordium is represented by a short, tapering, often comma-shaped rod or lamella which originates in the basal granule (figs. 22, 23, 26). As division progresses, the primordia increase in size (figs. 25, 28, 29). In more advanced stages (fig. 30), each of the new axostyles is clearly differentiated into the flattened expanded capitulum and the rod-shaped trunk. The posterior end of the trunk is typically blunt and not separated by a membrane from the surrounding cytoplasm. Further growth of the daughter axostyles is concerned mainly with the increase in length of the trunks by the addition of material at the distal ends.

The available material was unsuitable for the study of the development of the pelta.

The nucleus, situated about 1.5 microns below the anterior surface of the body, varies in shape, but it is usually ellipsoidal or ovoidal (figs. 1–12). A small nucleolus surrounded by a halo can be demonstrated in many nuclei. The nucleus, meas-

ured in 25 random organisms in a population of a strain from *Sauromalus obesus*, ranged 1.5–3 by 1.5–2, averaging 2.5 by 1.8. Similar measurements in a population of a strain from *Eumeces gilberti* gave a range of 1.5–2.5 by 1–1.5 and an average of 2 by 1.5.

All specimens stained with iron hematein (or hematoxylin) and all living organisms show a number of relatively large granules in the anterior third of the body. These granules are arranged usually in 3 (occasionally in 4) nearly parallel rows which encircle the nucleus (figs. 1, 8). Observations of living organisms indicate that the typical arrangement of the perinuclear granules is maintained only as long as the flagellates exhibit their normal activity. As the organisms start showing signs of degeneration, such as radical slowing down of the flagellar movement and a slight tendency toward amoeboid activity of the body, the granules become dispersed in the cytoplasm. This behavior suggests that in normal organisms the granules are kept in position by the relative rigidity of the peripheral part of the cytoplasm in which they are embedded.

Organisms maintained in culture may be seen to ingest solid food particles, such as grains of rice starch, by means of pseudopodia formed in the posterior part of the body. Occasionally, a flagellate ingests one or more members of its own species. Organisms that are easily identifiable can be found even in the cytoplasm of individuals in relatively advanced stages of division (figs. 24, 27). Also a number of specimens found in direct intestinal smears showed solid particles in their cytoplasm (figs. 8, 20). On several occasions large organisms were seen, the cytoplasm of which contained as many as 8 small and rounded flagellates that were clearly identifiable as *Proteromonas lacertae-viridis*. It is thus evident that *H. kirbyi* ingests solid food not only in culture, but also under natural conditions.

TAXONOMIC SUMMARY

Hexamastix kirbyi n. sp.

Type host.—Sauromalus obesus (Baird). Coachella Valley, 6 mi. E. Indio, Riverside Co., California, Syntype slides GP-132: 2, 3; GP-207: 3.

Additional hosts.—

Uma notata cowlesi Heifetz. Punta Peñasco, Mexico. Homoeosyntype slide GP-20: 3.

Sceloporus magister Hallowell. 4 mi. E. Blyth, Colorado River, Riverside Co., California. Homoeosyntype slide GP-96: 1.

Uta stansburiana Baird & Girard. 9 mi. E., 1 mi. N. Famoso, Kern Co., California. Homoeosyntype slides GP-92: 1, 2, 10.

Phrynosoma cornutum (Harlan). Clovis, Curry Co., New Mexico. Homoeosyntype slides GP-125: 1, 6, 7.

Eumeces gilberti Van Denburgh. 9 mi. E., 3 mi. N. Famoso, Kern Co., California. Homoeosyntype slides GP-75: 1, 2, 5, 6.

Diagnosis: (Measurements in microns.) Body typically ovoidal, measuring 8.5(5–17.5) by 7(3.5–15.5) in *Sauromalus obesus*, 8(5–12) by 6.5(2.5–10.5) in *Phrynosoma cornutum*, and 5.5(2.5–8.5) by 4(1.5–6) in *Eumeces gilberti;* five anterior flagella of unequal length ending in rods or knobs, longest flagellum measures in length 18.5(12–25.5) in *Sauromalus obesus*, 21(10.5–26) in *Phrynosoma cornutum*, and 13(12–15.5) in *Eumeces gilberti;* full complement of anterior flagella restored after cytoplasmic division, thus many 4-flagellate and some 3-flagellate immature organisms in actively dividing populations; recurrent flagellum with long acroneme, measures in length 17(13–22.5) in *Sauromalus obesus*, 20(12–25) in *Phrynosoma cornutum*, and 14(10.5–19) in *Eumeces gilberti;* parabasal apparatus situated dorsally and to the right of nucleus; parabasal body disk-shaped with darkly staining margin, larger bodies often show characteristic undulation that makes them appear somewhat bilobed; parabasal filament slender, applied to dorsal edge of disk; axostyle hyaline; capitulum of axostyle flattened expansion applied to left ventro-lateral surface of nucleus; trunk of axostyle slender rod (about 0.2 in diam-

eter), projects from posterior end of body for 5.5(2.5–8.5) in *Sauromalus obesus*, 6.5(2.5–11) in *Phrynosoma cornutum*, and 4(2.5–5.5) in *Eumeces gilberti*, distal part of projection tapers gradually or more abruptly to a point; pelta crescent-shaped membrane near anterior surface of body; nucleus ellipsoidal or ovoidal, with small nucleolus, about 1.5 below anterior surface of body, measures 2.5(1.5–3) by 1.8(1.5–2) in *Sauromalus obesus* and 2(1.5–2.5) by 1.5(1–1.5) in *Eumeces gilberti;* usually 3 (occasionally 4) parallel rows of relatively large perinuclear granules.

Hexamastix crassus n. sp.

(Figs. 33–42)

This new species was found in 4 species of lizards: the banded gecko, *Coleonyx variegatus* (2 out of 10); the iguanids, *Uta stansburiana* (1 out of 3) and *Phrynosoma cornutum* (1 out of 1); and the skink, *Eumeces gilberti* (1 out of 3).

All the individual hosts harbored several flagellate genera beside *Hexamastix*. *H. crassus* was the only species of this genus found in the gecko, but in the iguanids and in the skink it was accompanied by *Hexamastix kirbyi*.

DESCRIPTIVE ACCOUNT

(All measurements in microns.)

The body is ovoidal in shape (figs. 33–42). Fifty random organisms[2] ranged in length from 5 to 10 in width from 3.5 to 6.5, averaging 7 by 5.

In fully grown organisms there are 5 anterior flagella of unequal length (figs. 33, 35–37, 39, 40, 42). They originate in the basal granule complex (figs. 35, 39, 41), which appears often very large in iron hematein (or hematoxylin)-stained specimens (fig. 33). The flagella terminate in rods or knobs and are united in a short whip or column at their base (figs. 35, 39, 42). The longest anterior flagellum, measured in 25 random organisms, averaged in length 13(9.5–18.5).

Counts of the anterior flagella were made in 100 random specimens. Forty organisms had five, 30 organisms, four, and 10 specimens, only three flagella. The remaining 20 organisms were unsuitable for flagellar counts. The individuals with 3 flagella were the smallest in the population (fig. 38), and those possessing 4 anterior flagella also fell in the lower half of the size range (figs. 34, 41). While the available material was unsuitable for a study of morphogenesis, it is likely that the differences in the number and length of the anterior flagella may be explained by their behavior in division.

The recurrent flagellum, the diameter of which equals that of the anterior flagella, originates in the dorsal part of the basal granule complex (figs. 33, 35, 39); it terminates in a short acroneme. The flagellum, measured in 25 random specimens, ranged from 8.5 to 18.5 and averaged 12.5

The parabasal apparatus, as seen in specimens stained with protargol, is situated dorsally and to the right of the nucleus (figs. 36, 38, 39, 41). It consists of a short, distally curved, filament to which is applied a dorsally flattened discoid parabasal body (figs. 35, 36, 38, 39, 42). In many specimens, the entire body stains faintly with silver proteinate and appears to be homogeneous (figs. 34, 35, 37, 39), but in others a dark granule can be seen in the mid-dorsal part of the disk (figs. 36, 38, 41, 42).

The axostyle originates in the vicinity of the basal granule complex to which

[2] All measurements were made in one population of a strain from *Eumeces gilberti*.

it is connected (figs. 33–35, 39, 41). The anterior part of the axostyle is flattened and exhibits a certain amount of counterclockwise torsion (figs. 35–37, 39, 41). The flattened part extends into the posterior half of the body where the twist invariably becomes quite pronounced (figs. 33–42). Posterior to the twist, the axostyle becomes cylindrical and impregnates intensely with protargol (figs. 34–42). In many lateral views the flattened part is wider than the diameter of the cylindrical segment (figs. 33–36), resembling the capitular expansion of the more typical axostyles seen in numerous trichomonad species. The cylindrical part is relatively stout, averaging about 0.4 in diameter. A segment of this part projects from the posterior end of the body, and, some distance distal to it, tapers gradually (figs. 34, 41), or often more abruptly (fig. 35) to a fine filament. The terminal filament constitutes in many organisms about 40–50% of the total length of the axostylar projection, which, measured in 25 random organisms, averaged 5(2.5–7).

The pelta is a relatively slender crescent-shaped membrane situated near the anterior surface of the body (figs. 33–42). The membrane curves from the right-dorsal towards the left-ventral side of the body, ending near the left ventro-lateral surface of the nucleus (figs. 34, 36–38, 42). In some specimens the dorsal and ventral parts of the pelta taper gradually and terminate in fine filaments (figs. 34, 36–38, 42).

The nucleus, situated about 1 micron below the anterior surface of the body, is ellipsoidal (fig. 33) or ovoidal (figs. 34, 36–38, 42) in shape. It contains a small spheroidal nucleolus surrounded by a halo (fig. 33). Measurements of the nucleus in 25 random organisms gave a range of 1.5–3 by 1–1.5, and an average of 2 by 1.5.

All well-preserved specimens stained with iron hematein (or hematoxylin) show usually 4 rows of perinuclear granules (figs. 33, 33a). The granules range in size from very small to relatively large.

TAXONOMIC SUMMARY

Hexamastix crassus n. sp.

Type host.—Eumeces gilberti Van Denburgh. 9 mi. E., 3 mi. N. Famoso, Kern Co., California. Syntype slides GP-75: 1, 2, 5.

Additional hosts.—

Coleonyx variegatus (Baird). Sentenac Canyon, San Diego Co., California. Homoeosyntype slide GP-87: 1; 7 mi. S. Vidal, Riverside Mts., Riverside Co., California. Homoeosyntype slides GP-89: 2, 3, 6.

Uta stansburiana Baird & Girard. 9 mi. E., 1 mi. N. Famoso, Kern Co., California. Homoeosyntype slides GP-92: 1, 2, 11.

Phrynosoma cornutum (Harlan). Clovis, Curry Co., New Mexico. Homoeosyntype slides GP-125: 1, 6.

Diagnosis (from *Eumeces gilberti*) (Meaurements in microns.): Body ovoidal, measuring 7(5–10) by 5(3.5–6.5); in fully developed organisms five anterior flagella of unequal length ending in rods or knobs, longest flagellum measures in length 13(9.5–18.5); recurrent flagellum with short acroneme, measuring in length 12.5(8.5–18.5); parabasal body dorsally flattened disk with dark mid-dorsal granule, dorsal part of disk applied to fine parabasal filament; anterior part of axostyle flattened, with certain amount of counterclockwise torsion, extending into posterior part of body; posterior part of axostyle cylindrical (about 0.4 in diameter), projects from posterior end of body for 5(2.5–7), distal part of projection tapers gradually or more abruptly to a long filament; pelta slender crescent-shaped membrane near anterior surface of body; nucleus ellipsoidal or ovoidal, with small spheroidal nucleolus, about 1 below anterior surface of body, measures 2(1.5–3) by 1.5(1–1.5); usually 4 rows of perinuclear granules of varying size.

DISCUSSION

The differences between the two, at times coexistent, forms of *Hexamastix* from lizards are sufficiently great to justify their specific separation. The most important basis for this separation is found in the structure of the parabasal apparatus and in that of the axostyle. In fact, the latter organelle in *Hexamastix crassus* is in some respects different from the axostyles found in all other members of the genus *Hexamastix*.

Six species of *Hexamastix* have been described from termites (see Kirby, 1930), one each from European mole crickets (see Grassé, 1926), crane-fly larvae (see Ludwig, 1946), and amphibians (see Honigberg and Christian, MS.), and 3 from rodents (see Kirby and Honigberg, 1949 and Nie, 1950). My observations of 10 of these species and the examination of descriptions of the remaining 2 [*Hexamastix gryllotalpae* Grassé and *Hexamastix tipulae* (Ludwig)] indicate that while some of the 12 specific names may represent synonyms, all of the hitherto reported 6-flagellate monocercomonads can be readily differentiated from *H. kirbyi* and *H. crassus* on valid morphological grounds.

There is much evidence that many species of flagellates inhabiting the intestines of squamate reptiles are morphologically indistinguishable from those found in amphibians. However, the members of the genus *Hexamastix*, harbored by lizards, differ in many characters from *Hexamastix batrachorum* (Alexeieff) of salamanders. The most significant differences occur in the degree of development of the pelta, which is considerably larger in the flagellate from amphibians, and in the shape of the parabasal body which, "V"-shaped in *H. batrachorum*, is disk-shaped in both species from lizards. Also the axostyle of the organism from salamanders is stouter than that of *H. kirbyi* and quite different in structure from the homologous organelle in *H. crassus*.

As far as can be ascertained, no extensive study of division has been hitherto made in any species of *Hexamastix*. According to Kirby (1930), in *Hexamastix claviger* from termites, 2 anterior and 1 recurrent flagella were associated with each of the daughter mastigonts in early division stages, and the full flagellar complements were restored prior to the division of the parental nucleus. Such behavior of the flagella in division is different from that observed by me in *H. kirbyi*. However, it is possible that, in the few living organisms on which his study of early divisions of *H. claviger* was based (personal communication received by me from Dr. Kirby in 1951), he miscounted the number of rapidly moving anterior flagella. The new recurrent flagellum might have achieved considerable length relatively early in division, even before any new anterior flagella appeared. Thus one would expect to find 3 anterior flagella and the new recurrent flagellum associated with 1 daughter basal granule complex, and 2 anterior flagella and the old recurrent flagellum with the other. Since the exact time of the outgrowth of the several new flagella and that of the restoration of the full flagellar complements in the daughter mastigont systems varies widely among different trichomonad species, the differences in time of appearance of the new flagella in *H. claviger* and *H. kirbyi* are not surprising.

In *Hexamastix termopsidis*, Kirby (1930) observed significant variations in the number of the anterior flagella in solitary organisms that otherwise showed no signs of division. He suggested that probably the anterior flagella "separate three and two

in division, and that the outgrowth to completion takes place slowly, and one flagellum at a time." Thus in this species the anterior flagella appear to behave very much like those of *H. kirbyi*. The same is true of *H. tipulae* in which Ludwig (1946) observed each mastigont to receive 3 of the parental flagella, 2 anterior and 1 recurrent going to 1 daughter and 3 anterior to the other. Furthermore, he could see only 3 flagella associated with each basal granule complex even in organisms that were in the process of cytokinesis. There is little doubt that the differences in the number of anterior flagella reported by Nie (1950) in *Hexamastix caviae* and *Hexamastix robustus* depended upon the time of appearance of these organelles in division.

The behavior of all the mastigont organelles of *H. kirbyi* in division agrees in most respects with that reported from other trichomonad genera in which morphogenesis has been studied (for recent reviews see Kirby, 1944 and Honigberg, 1951).

On the basis of information gathered from investigations of other trichomonads it could have been predicted that in *H. kirbyi* the new recurrent flagellum would appear quite early in division and reach its full development prior to completion of the nuclear division. Similarly it could have been anticipated that the outgrowth of the new recurrent flagellum would precede that of the new anterior flagella. However, while in some instances the former appeared a short time before any of the latter, in others the new recurrent and 1 of the new anterior flagella seemed to grow out almost simultaneously, relatively late in division, each in 1 of the 2 daughter mastigonts.

While the general pattern of morphogenesis of the several mastigont organelles is the same in all trichomonads, certain significant differences exist among the various genera and species. These differences involve most often the time of outgrowth of the new flagella and the details of behavior of the parental parabasal apparatus. The more information is gathered about morphogenesis of trichomonads, the clearer it becomes that similarities in behavior of the mastigont organelles in division bring out the phylogenetic relationships among the species.

SUMMARY

Two species of *Hexamastix* (Alexeieff) were found in 9 lizards belonging to 7 genera and as many species. Three iguanid species were infected only with **Hexamastix kirbyi** n. sp., the banded geckos only with **Hexamastix crassus** n. sp.; the 2 remaining species of iguanids and the skink harbored both species of this flagellate genus.

Hexamastix kirbyi has a typically ovoidal body. Fully developed organisms possess 5 anterior flagella of unequal length, which end in rods or knobs. The single recurrent flagellum about equals the anterior flagella in length and ends in a relatively long acroneme. The outgrowth of the new flagella takes place relatively late in division, and the full flagellar complement is restored after cytokinesis. Thus some 3- and many 4-flagellate organisms are found in actively dividing populations. The parabasal apparatus consists of a disk-shaped body, that often shows a typical undulation, and of a fine filament. Part of the parental parabasal is discarded early in division. One daughter receives the remaining part of the old organelle, while the other develops its own parabasal apparatus. The slender hyaline axostyle is expanded anteriorly into a flattened capitulum. The capitulum, applied to the ventrolateral surface of the ellipsoidal or ovoidal nucleus, remains in a close spatial relationship with the crescent-shaped pelta. During early stages of division the old

axostyle and pelta are discarded, and new organelles develop in each daughter mastigont. The organism contains usually 3 rows of large perinuclear granules.

Hexamastix crassus is characterized by an ovoidal body. Fully developed organisms have 5 anterior flagella of unequal length which end in knobs or rods. The single recurrent flagellum nearly equals the anterior flagella in length and ends in a short slender filament. The parabasal body is a dorsally flattened disk which contains a darkly staining granule. The dorsal part of the disk is applied to a slender filament. The anterior flattened part of the axostyle, probably continuous with the slender crescent-shaped pelta, exhibits a counterclockwise torsion. The posterior part of the axostyle is cylindrical. Its terminal segment projects from the body and tapers to a long fine filament. The ellipsoidal or ovoidal nucleus is surrounded by 4 rows of granules which vary in size.

Literature Cited

Balamuth, W. and Sandza, J. G. 1944 Simple standardized culture medium for phsiological studies on *Entamoeba histolytica*. Proc. Soc. Exper. Biol. and Med. **57:** 161–163.

Grassé, P. P. 1926 Contribution a l'étude des flagellés parasites. Arch. Zool. Exper. et Gén. **65:** 345–602.

Honigberg, B. M. 1951 Structure and morphogenesis of *Trichomonas prowazeki* Alexeieff and *Trichomonas brumpti* Alexeieff. Univ. Calif. Publ. Zool. **55:** 337–394.

——— and Christian, H. H., MS. Characteristics of *Hexamastix batrachorum* (Alexeieff). (In press).

Kirby, H. 1930 Trichomonad flagellates from termites. I. *Tricercomitus* gen. nov. and *Hexamastix* Alexeieff. Univ. Calif. Publ. Zool. **33:** 393–444.

———1944 Some observations on cytology and morphogenesis in flagellate protozoa. J. Morph. **75:** 361–421.

——— and Honigberg, B. 1949 Flagellates of the caecum of ground squirrels. Univ. Calif. Publ. Zool. **53:** 315–366.

Ludwig, F. W. 1946 Studies on the protozoan fauna of the larvae of the crane-fly, *Tipula abdominalis*. I. Flagellates, amoebae, and gregarines. Trans. Amer. Micr. Soc. **65:** 189–214.

Nie, D. 1950 Morphology and taxonomy of the intestinal protozoa of the guinea-pig, *Cavia porcella*. J. Morph. **86:** 381–494.

Explanation of Plates

All figures were made with the aid of camera lucida, unless otherwise stated, at the original magnification of ×4200, from preparations fixed in Bouin's fluid (Bou.), Hollande's cupric picroformol (Holl.), and Schaudinn's fluid (Sch.). Unless noted otherwise, the preparations were treated with protargol. The figures were reduced in print by about one-third.

Abbreviations: a.fl., anterior flagella; ax., axostyle; pb., parabasal body; pe., pelta; r.fl., recurrent flagellum.

Plate I

Hexamastix kirbyi n. sp.

Figs. 1–7 from *Sauromalus obesus;* figs. 8–11 from *Eumeces gilberti;* figs. 12–17 from *Phrynosoma cornutum.*

Figs. 1, 8 represent organisms stained with iron hematein.

Fig. 1. Right view showing all the mastigont organelles except for the parabasal apparatus. Note the large basal granule complex and the 3 rows of perinuclear granules. Bou.

Fig. 2. Left view of the anterior part of the body of an organism with 4 anterior flagella. Holl.

Fig. 3. Left view of a young organism with 3 anterior flagella. Note the small parabasal, the relationship of the pelta to the anterior part of the axostyle, and the long acroneme of the recurrent flagellum. Holl.

Fig. 4. Right view. The pelta shows clearly. The parabasal failed to stain. Holl.

Fig. 5. Right view of the anterior part of the body. The pelta is quite large; the parabasal body is a simple disk. Holl.

FIG. 6. Left view of the anterior part of the body of a large organism. Note the large parabasal body which shows the typical undulation and thus appears bilobed. Holl.

FIG. 7. Left-ventral view showing all the mastigont organelles. Observe the capitular expansion of the axostyle, the large pelta, and the long acroneme of the recurrent flagellum. Holl.

FIG. 8. Left view. Note the 2 granules in the basal granule complex and the 3 rows of perinuclear granules. Sch.

FIG. 9. Right view. The pelta is large. The large parabasal body is associated with a fine parabasal filament. Holl.

FIG. 10. Dorsal view. One anterior flagellum is much shorter than the remaining 4. Holl.

FIG. 11. Right view showing a typical arrangement of all the mastigont organelles. Holl.

FIG. 12. Right view of a large organism, with a saddle-shaped parabasal body and a relatively wide capitular expansion of the axostyle. Holl.

FIGS. 13–17. Diagrams of parabasal bodies. Holl.

PLATE II

Hexamastix kirbyi n. sp.

FIG. 18 from *Phrynosoma cornutum;* fig. 20 from *Uta stansburiana;* figs. 19, 21–25 from *Sauromalus obesus.* All figures, Holl.

FIG. 18. The discarded part of the parental parabasal body is still in close proximity to the retained portion.

FIG. 19. The pelta shows signs of degenerative hypertrophy. One of the small parabasals is new; the other represents the retained portion of the parental organelle. The discarded part of the old parabasal is seen in the cytoplasm.

FIG. 20. Two anterior flagella and the recurrent flagellum as well as 1 parabasal are associated with 1 daughter basal granule complex, while 3 anterior flagella and the second parabasal with the other. A large inclusion is seen in the cytoplasm.

FIG. 21. The components of the 2 daughter mastigonts are as in fig. 20, but the basal granules are further apart. Only the old pelta and a part of the capitulum of the old axostyle are missing.

FIG. 22. Most organelles are as in fig. 21, but the capitulum of the old axostyle has already degenerated and the parabasals have increased in size. Primordia of new axostyles are associated with each of the daughter basal granule complexes, which are connected by the extranuclear spindle.

FIG. 23. Most organelles are as in fig. 22, but only a very small segment of the old axostyle still remains and the new parabasals are quite large.

FIG. 24. Most organelles as in fig. 23, but the remnant of the old axostyle is no longer present. Note a partly digested flagellate in the cytoplasm of the dividing organism. Orig. magn. ×2800.

FIG. 25. Relatively advanced stage of division with daughter nuclei a considerable distance apart and large primordia of the new axostyles. The flagellar complement is still as in figs. 20–24.

PLATE III

Hexamastix kirbyi n. sp.

All figures are from *Sauromalus obesus.* Holl.

FIGS. 27–29, 32 were drawn at the original magnification of ×2800.

FIG. 26. Nuclear division is not yet completed. A new short recurrent flagellum is present in the mastigont to the observer's right.

FIG. 27. A giant organism with large parabasals. The nuclear division is completed. A stump of a new anterior flagellum is present in the mastigont to the observer's left. Two partially digested flagellates are seen in the cytoplasm of the dividing form.

FIG. 28. Daughter nuclei are far apart. Short stumps of new flagella are associated with each mastigont system.

FIG. 29. A giant organism in a stage of division similar to that in fig. 28. Each daughter mastigont contains a short new flagellum. Note the very large parabasals and the new axostyles.

FIG. 30. Late division. Four flagella are associated with each daughter mastigont. The new axostyles are differentiated into the capitulum and the still incomplete trunk.

FIG. 31. Probably a stage just preceding cytokinesis (or, because of the absence of extranuclear spindle, a young double individual). The flagellar complement is as in fig. 30, but all the other organelles in both mastigonts are fully developed.

FIG. 32. Double organism with 2 fully developed mastigonts, each containing only 4 anterior flagella.

PLATE IV

Hexamastix crassus n. sp. from *Eumeces gilberti*

FIGS. 33, 33a, Sch.; figs. 34–42, Holl.

FIG. 33. Right view showing all the mastigont organelles, except for the parabasal apparatus. Note the large basal granule complex and the four parallel rows of perinuclear granules. Iron hematein. Orig. magn. ×5600.

FIG. 33a. Diagram of the left view of the anterior part of the organism represented in fig. 33 showing the rows of perinuclear granules. Orig. magn. ×5600.

FIGS. 34 and 38. Right views of immature organisms, with 4 and 3 anterior flagella respectively. Note the long filamentous terminal segment of the axostyle in fig. 34.

FIG. 35. Left view. One of the anterior flagella is much shorter than the remaining 4. Note the relationship of the pelta to the anterior part of the axostyle.

FIGS. 36 and 40. Right views. In fig. 36 the axostyle shows clearly the typical, but somewhat exaggerated, counterclockwise torsion. In fig. 40 observe the relatively wide pelta and the stout axostyle.

FIGS. 39 and 41. Dorsal views. In both figures note the origin of the flagella and parabasal apparatus in the basal granule complex, as well as the relationship of the pelta to the axostyle. The torsion of the axostyle is greatly exaggerated in fig. 39.

FIG. 42. Right-ventral view. Note the typical disk-shaped parabasal body with the middorsal granule, the parabasal filament, and the pelta, the dorsal part of which tapers gradually to a fine filament.

PLATE I

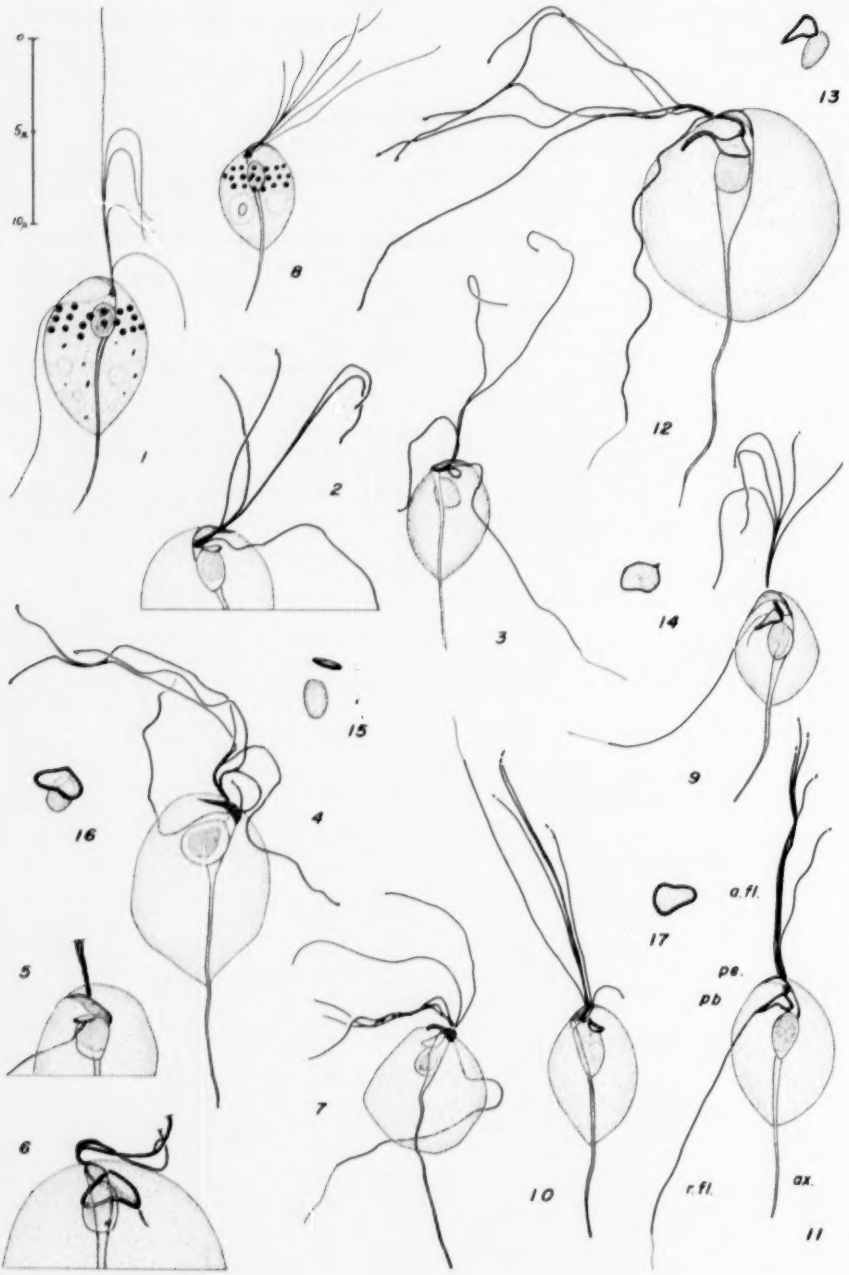


PLATE II

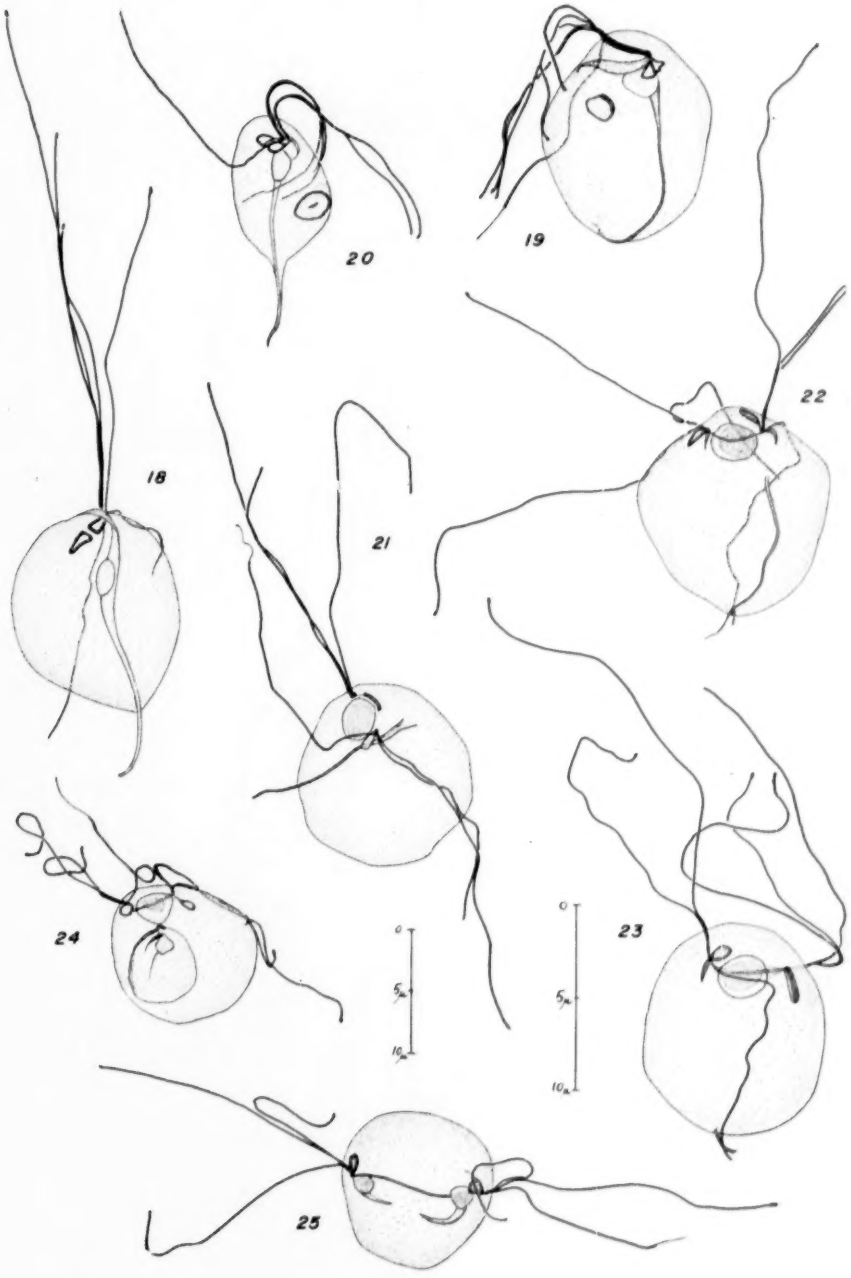


PLATE III

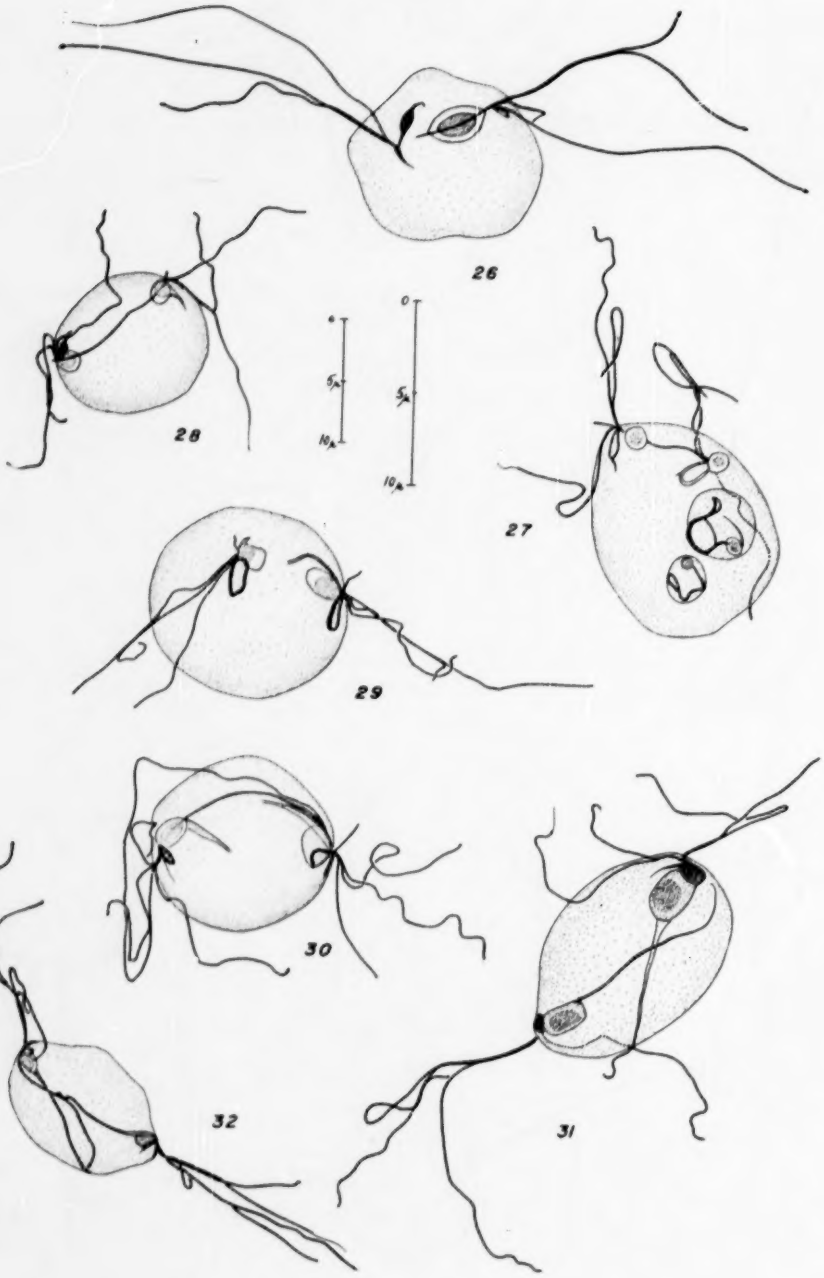
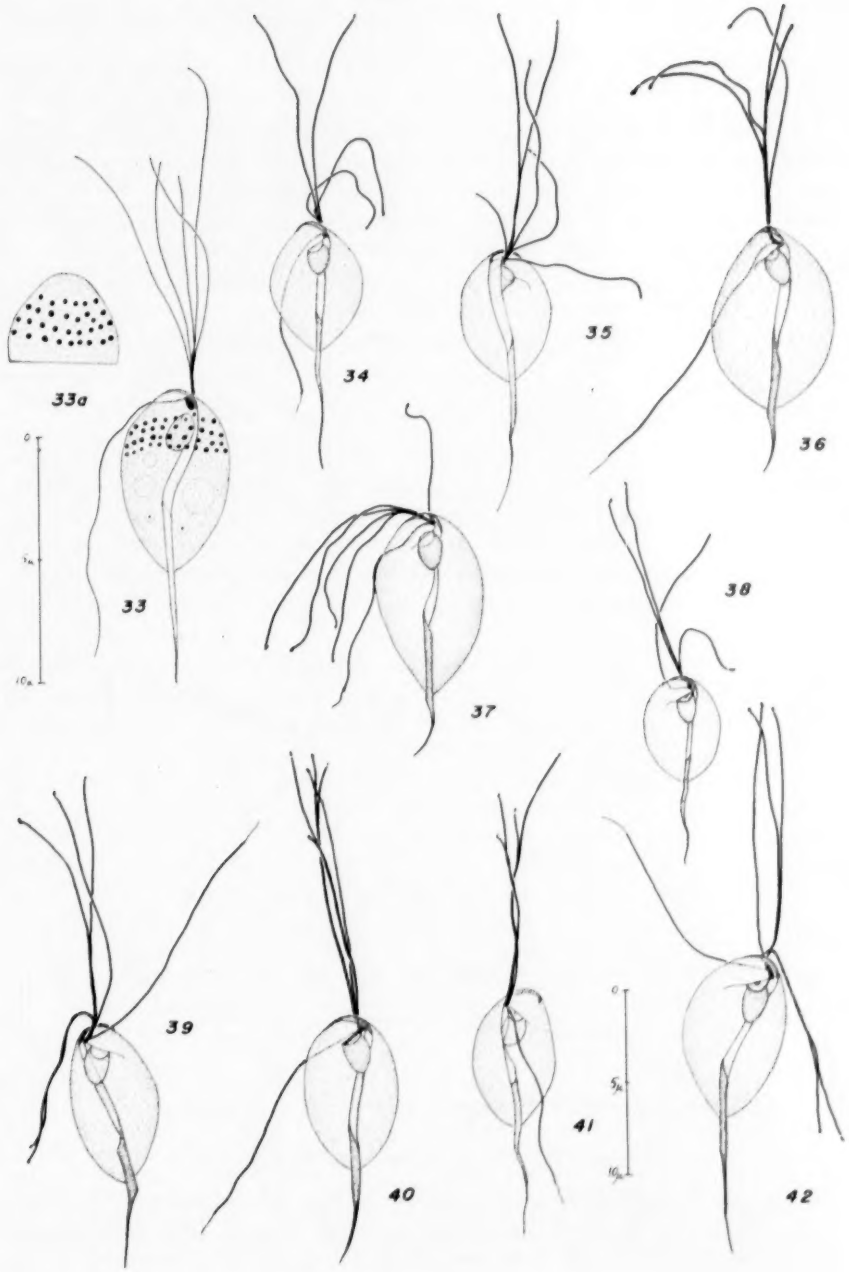


PLATE IV



IXODES KOHLSI, A NEW SPECIES OF TICK FROM AUSTRALIA

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In December 1953 I received, from Mr. Glen M. Kohls of the Rocky Mountain Laboratory, Hamilton, Montana, U.S.A., four lots of ticks from *Eudyptula minor* for further investigation. The material in question was as follows:

RML 21545. Julia Percy Island, Australia, January 1936, 4 males, 3 females, and 6 nymphs, *Ixodes percavatus* Neumann. J. A. Tubb.

RML 21547. Sir Joseph Banks Island, Australia, January 1937, 6 females, *I. percavatus* Neumann. J. A. Tubb.

RML 13904. Robe, S. Australia, 1 female, *I. eudyptidis* Maskell, det. D. C. Swan.

RML 27589. Tolaga Bay near East Cape in North Island, New Zealand. 1934, 1 female, *I. eudyptidis*, det. L. J. Dumbleton.

The first three lots are one species which bears a general resemblance to *Ixodes rothschildi* Nuttall and Warburton, 1911 (Arthur, 1953) but is consistently different from the latter in the dentition of the hypostome, the form of the tarsi, and the structure of Haller's organ. Haller's organ can be used to separate these lots from RML 27589 as the latter has an open posterior "capsule" (Figs. 1A and B); in

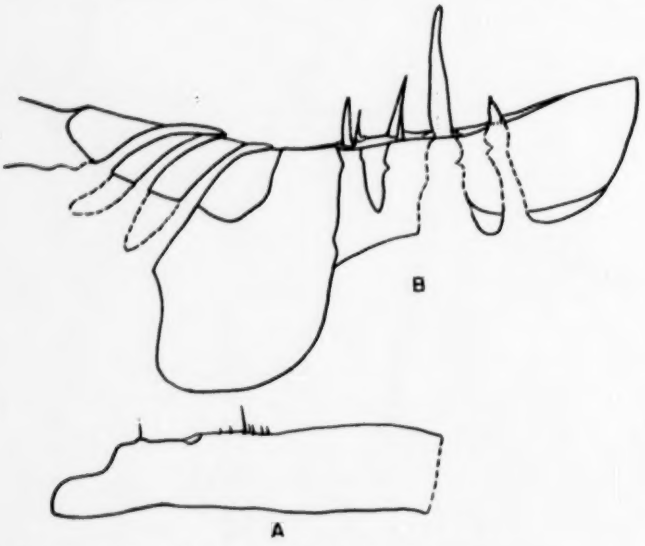

FIG. 1. *Ixodes eudyptidis* Maskell. A. Tarsus I. B. Haller's organ.

the other lots the capsule is closed except for a small antero-dorsal aperture (Fig. 2I). RML 27589 agrees well with *Ixodes eudyptidis* Maskell, 1885, as determined by Dumbleton (1953), and there can be little doubt about the correctness of his diagnosis.

The following data are relevant to lots RML 21545, RML 21547, and RML 13904. As there is substantial similarity between the females of this species and *I. rothschildi*, only the differences will be considered.

Description of female

Hypostome (Fig. 2). Length 0.41–0.44 mm., breadth 0.16 mm. Broad, gently rounded distally. Denticles arranged from distal to proximal end as one row of 6/6, one row of 5/5, two rows of 4/4, six rows of 3/3, five-six rows of 2/2 extending to the base. This arrangement is remarkably uniform in the forms examined; a median ventral unarmed area extends almost the whole length of the hypostome.

Anal grooves constricted behind the middle, then becoming parallel-sided; in fully fed specimens becoming more or less parallel-sided except for a slight notch immediately behind the anus. In *I. rothschildi* the anal groove is parallel-sided without evidence of constriction.

Genital aperture located between coxae II, or the intercoxal space between coxae II and III.

Tarsi. These taper more abruptly than do those of *I. rothschildi* which is emphasized by comparing Figs. 3A and 3B, and of *I. rothschildi* (Fig. 7) as shown in an earlier paper (Arthur, 1953).

Haller's organ. In *I. rothschildi*, this is an open structure (Arthur, 1953, Fig. 8), but in the present species it is closed (Fig. 21).

The constancy of these structural differences suggests that these specimens are to be considered as a distinct species.

This species is named for Glen M. Kohls, of the Rocky Mountain Laboratory, in recognition of his numerous contributions to our knowledge of ticks, and as a measure of respect and esteem for his unfailing and generous assistance to all tick workers.

Specimens of lot RML 21545 consisted of 3 females, 6 nymphs and 4 males. The contained legend in the tube reads "*I. percavatus* Neum., from *Eudyptula minor*. Julia Percy Island, Australia. January 1936. J. A. Tubb." That the specimens were removed from the host appears to be doubtful; rather it would seem that they were collected from the nesting sites of the bird, as nymphal development is very advanced. The nymphal cuticle surrounds the adult form which is fully developed externally except for the straightening of the downwardly-bent capitulum and the stretching of the legs. Examination of the developing males and females removed from the nymphal exuviae reveals that the nymphs, males, and females belong to one species. The males of this collection are of some interest in that specimens removed from the nymphal skin are dark brown in color, as also are two of the four specimens in the collection. The other two, whilst obviously of the same species, do show varying degrees of sclerotization of certain plates, as will be detailed later. The form of the hypostome of the male relates these ticks to species of which the females only have been found on the host (Nuttall, 1911) and gives additional support to the suggestion that they were collected from the nesting sites of their hosts.

Description of male

Body. Oval, widest at about middle. Color dark brown. Marginal fold beginning opposite coxa I in two specimens (Fig. 17) (where sclerotization of plates is irregular) and about opposite coxa III (Fig. 16) (in the specimens where sclerotization of plates is more regular and in males removed from nymphal skins). Where the marginal fold is clearly defined, there are four or five heavily sclerotized spots at about the level of second intercoxal space.

Capitulum (Figs. 12, 13, 15). Basis more or less rectangular with lateral margins straight. Cornua absent. Length 0.5 mm. Palpi short and very broad. Bristles numerous along the margins with a median ventral strong row on articles II and III (Fig. 13). Suture line between articles II and III indistinct. Dorsal surface of palp quite strongly curved so that projecting

article IV is directed antero-ventrally (Fig. 15). Length of articles II and III 0.36 mm.; maximum breadth 0.18 mm. (Figs. 12 and 13). Dorsal ridge of basis sinuous and elevated (Figs. 12 and 15) and bearing two irregularly defined patches of pores (about 9–11 pores per patch). Basis continued back beyond dorsal ridge as a parallel-sided plate terminating in a broadly rounded margin. Lower side of basis shorter than upper and having parallel sides (Fig. 13). Palpiger bearing article I projects out from the side.

Hypostome. Short and broad, indented apically (Fig. 14). Denticles limited to serrations marginally except for two teeth on outer margin distally. Length 0.25 mm., breadth 0.18 mm.

Scutum. Smooth, shiny, more strongly convex in front; postero-lateral margins less well defined in males removed from the nesting site (?) with a strong groove marginally. Whether this depression on the scutum in the four males collected from the nest (?) is a natural phenomenon or a product of fixation cannot at present be stated. Males removed from nymphal skins do not show this groove. Punctations small and numerous, more numerous in lateral and posterior areas. In specimens removed from alcohol, the degree of sclerotization varies as shown in Figs. 16, 17, and 18, but if allowed to dry to some extent these areas of weaker sclerotization are no longer evident. They reappear however if resoaked in alcohol. The distinction between the scutum and the marginal body fold is ill defined in the male about to emerge from the nymphal cast. Cervical grooves short, broad, deep, divergent; limits of pseudoscutum faintly indicated. Length 3.5 mm., breadth 2.5 mm. Hairs few and small, often absent. Scapulae short, rounded, emargination distinct but shallow, with interscapular portion of scutum projecting forward and broadly rounded. No lateral carinae.

Ventral plates (Fig. 19). Pregenital plate pentagonal with V-shaped incision anteriorly, supported antero-laterally by two crescent-shaped sclerites at base of coxa I. Small transverse rectangular post-genital plate. Median plate irregularly sclerotized posteriorly as indicated in Figs. 19A, B, C; longer than adanal plates. Adanal plates roughly triangular in outline, with mesal face frequently irregular in profile. Intersegmental membrane appears to be well developed in two specimens, but this may be due to the irregular sclerotization of the plates. Anal plate horseshoe-shaped, shorter than adanal plates usually; sclerotization irregular. Short white hairs, points of insertion of hairs dark brown.

Legs (Figs. 20A, B). Strong, long and thick; well supplied with long, strong bristles. Tarsi narrowed abruptly subapically and directed downward. Length of tarsus I 0.68 mm., length of tarsus IV 0.82 mm. Haller's organ (Fig. 21) a closed capsule, as in female and nymph.

Coxae. Convex, smooth, shining with few short white hairs. Absence of spurs. Increase in size from coxa I to IV (Fig. 19A).

Spiracular plate round with macula near center or antero-ventrally.

Genital aperture (Fig. 19A). In intercoxal space between coxae II and III.

Anus. Located well back on opisthosoma; anal grooves, horseshoe-shaped.

Nymph

Capitulum (Fig. 6). Length, tip of cornua to tip of hypostome, 0.37–0.39 mm., greatest width 0.27–0.28 mm. Basis broad, lateral margins short, more or less parallel-sided. Cornua definite as rounded protuberances, short (Fig. 5). Surface a little convex, smooth, impunctate. Palpi gently curved laterally and more so medially; combined length of articles II and III, 0.23 mm., width 0.063 mm. Suture line between articles II and III indistinct. Surface smooth, few short hairs. Basis in ventral view rounded behind, surface smooth and impunctate; auriculae as rounded, lateral salient lobes.

Hypostome (Fig. 7). Much as in female; denticles from base to tip in five rows 2/2, six rows of 3/3, and one row of 4/4. Length 0.25 mm.

Scutum (Fig. 8). Length 0.61 mm., width 0.49 mm. across the shoulders; widest in front of the middle. Surface shining, few punctations, moderately long white hairs. Lateral carinae absent. Cervical grooves long, shallow, reaching well back to postero-lateral margins.

Coxae as in female (Fig. 9).

Spiracular plate (Fig. 11). Small, elongate oval; goblets few. Long axis arranged transversely to corresponding body axis. Length of longer axis 0.14 mm.

Holotype—female from RML 21545.

Allotype—male from RML 21545.

Paratypes—two males (since a total of only three are now available in the collection) and two females from RML 21545; six females from RML 21547; one female from RML 13904.

Holotype female, allotype male, and female paratypes are deposited at the Rocky Mountain Laboratory. Male and female paratypes deposited in the British Museum (Natural History) and in the U. S. National Museum.

ACKNOWLEDGMENT

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REFERENCES CITED

ARTHUR, D. R. 1953 The systematic status of *Ixodes percavatus* var. *rothschildi* Nuttall and Warburton, 1911. Parasitology **43**: 222–226.

DUMBLETON, L. J. 1953 The ticks (Ixodoidea) of the New Zealand sub-region. Cape Expedition Bulletin No. 14, pp. 1–28, plus plates 1–4.

NUTTALL, G. H. F. 1911 On the adaptation of ticks to the habits of their hosts. Parasitology **4**: 46–67.

EXPLANATION OF PLATE I

FIGS. 2–14. *Ixodes kohlsi* n. sp. FIG. 2. Female. Hypostome. FIG. 3A, B. Female tarsus I and IV. FIG. 4. Female. Scutum. FIG. 5. Nymph. Basis capituli showing small blunt cornua (dorsal view). FIG. 6. Nymph. Basis capituli showing auriculae (ventral view). FIG. 7. Nymph. Hypostome. FIG. 8. Nymph. Scutum. FIG. 9. Nymph. Coxae I to IV. FIG. 10. Nymph. A. Tarsus I. B. Tarsus IV. FIG. 11. Nymph. Spiracle. A, anterior; V, ventral. FIG. 12. Male. Basis capituli (dorsal view). FIG. 13. Male. Basis capituli (ventral view). FIG. 14. Male. Hypostome; note that second file is limited to two teeth distally.

EXPLANATION OF PLATE II

FIGS. 15–21. *Ixodes kohlsi* n. sp. FIG. 15. Male. Side view of basis capituli; note the salient dorsal ridge, and the downwardly curving portion of the palp. FIG. 16. Dorsal view of less irregularly sclerotized male. The heavy stippling in the postero-lateral region represents the less sclerotized portions of the scutum. It also represents the approximate distribution of the "groove." FIG. 17. Dorsal view of more irregularly sclerotized male. Note the "spots" between coxae II and III; the wider body fold when compared with FIG. 16; the upward extensions of the adanal, anal, and genital grooves. The heavy stippling denotes the same features as in FIG. 16. FIG. 18. Sketch of the scutum of another irregularly sclerotized male. FIG. 19. A. Ventral view of male to show arrangement and relative size of plates. B and C. Patterns of irregularly sclerotized ventral plates. The stippling represents the more heavily sclerotized and pigmented portions. FIG. 20 A, B. Male. Tarsus I and IV. FIG. 21. Male. Haller's organ. Except for size this organ is similar in the female. Compare with FIG. 1 and with FIG. 8 of Arthur (1953). The figures 1, 2a, 2b, 3a, 3b, 4, and 5 refer to the successive appearance of the sensilla as seen in optical section when viewing from the outer side of the capsule to the inner The letters a and b refer to the simultaneous appearance of two sensilla in the same field of vision.

PLATE I

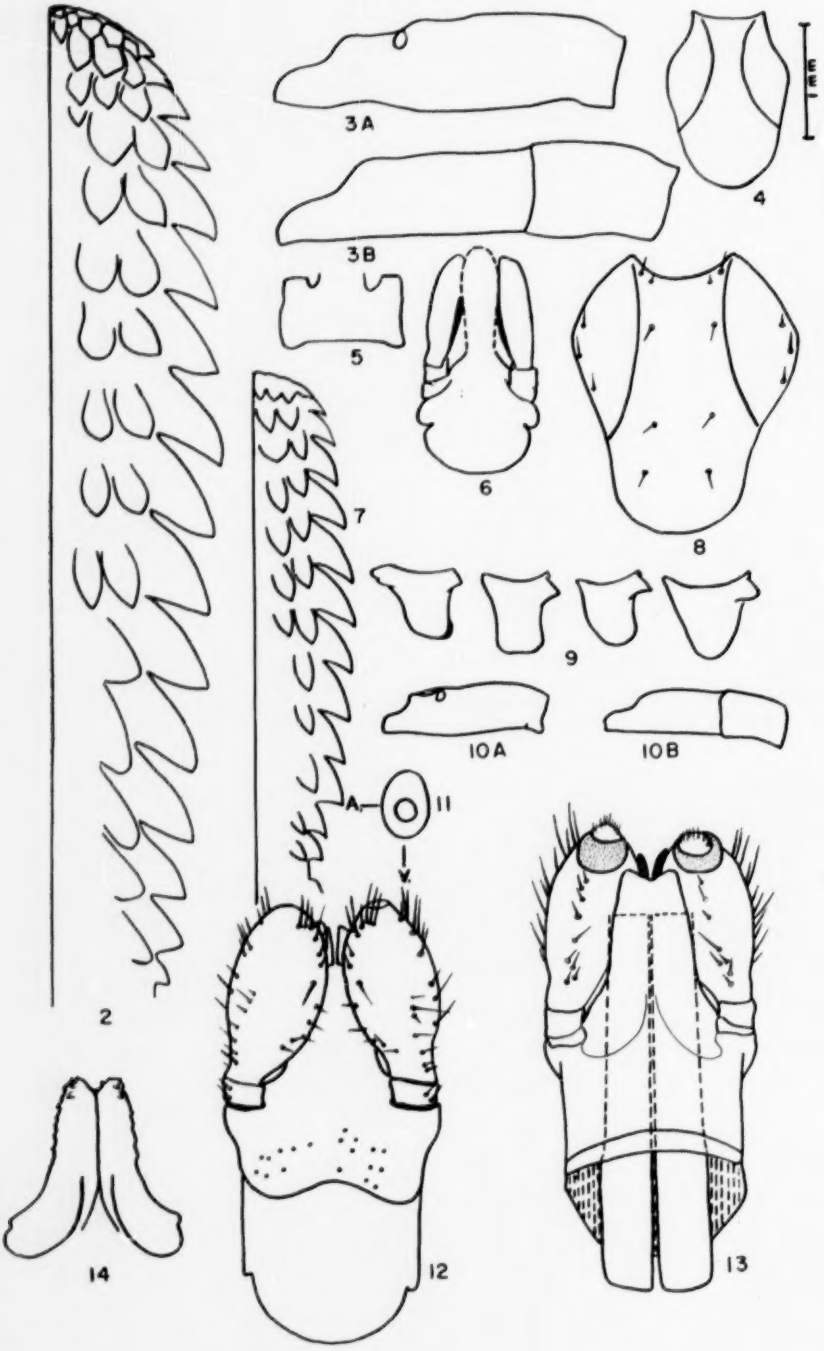
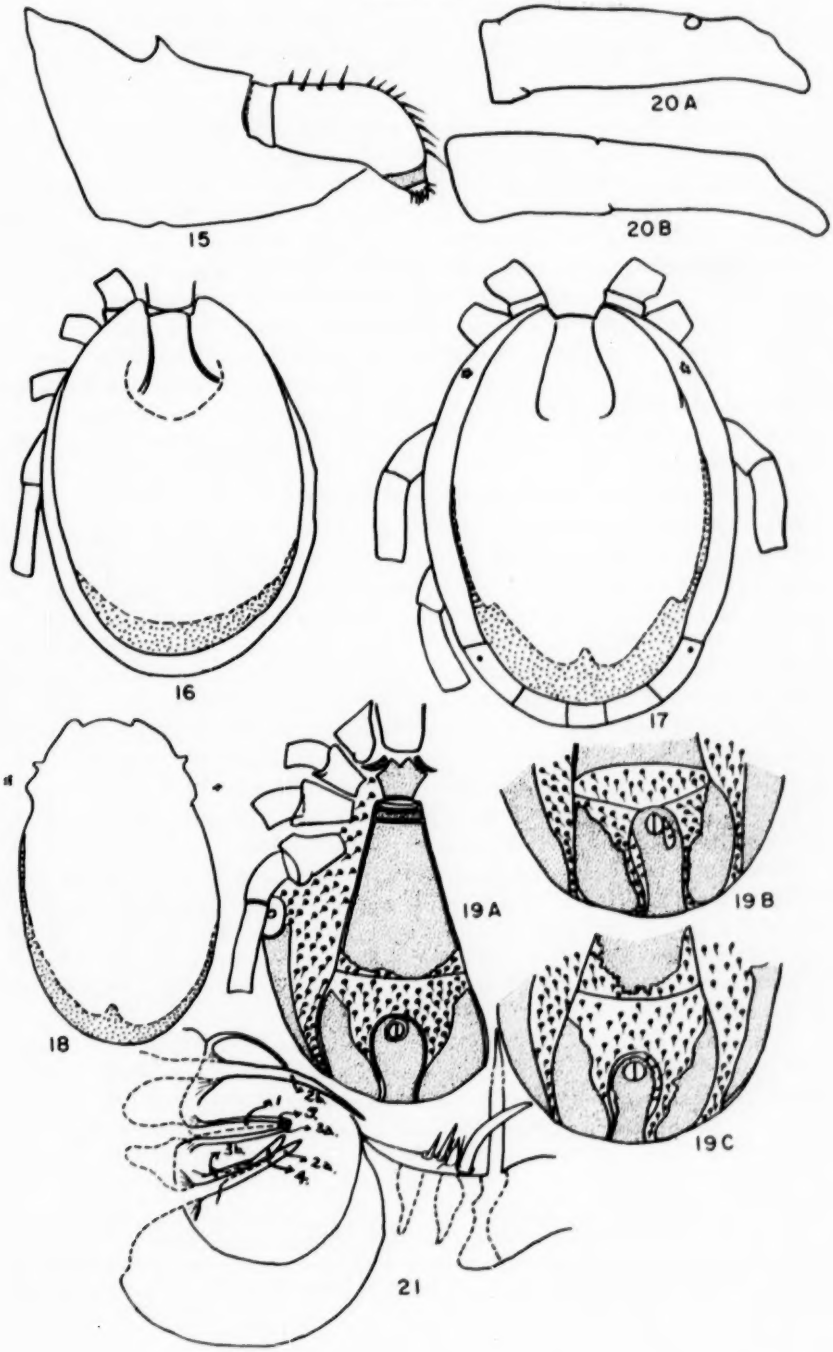


PLATE II



GERMINAL DEVELOPMENT IN THE SPOROCYSTS OF A BIRD SCHISTOSOME *TRICHOBILHARZIA PHYSELLAE* (TALBOT, 1936)[1]

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INTRODUCTION

The cercaria of *Trichobilharzia physellae* was described by Talbot (1936) from *Physa parkeri* Currier from Douglas Lake, Michigan, near the University of Michigan Biological Station. Several other species of *Physa* are also known to serve as intermediate hosts for this species. The adult has been reported in natural infections in the blue-winged teal (McLeod and Little, 1942) and from experimental infections in canaries, pigeons and domestic ducks (McMullen and Beaver, 1945). The cercariae of this species have been shown to produce schistosome dermatitis in both natural and experimental infections (Cort, 1950). A preliminary report of some of our results was published in an earlier paper (Cort, Ameel and Van der Woude, 1954a).

MATERIALS AND METHODS

The larval trematode infections in *Physa parkeri* in the Douglas Lake region have been studied by Cort, Olivier and McMullen (1941) and some of their results were helpful in this investigation. They found the cercaria of *T. physellae* in this snail in the early summer in three of their collecting areas on Douglas Lake, viz., the South Fishtail Bay Area, the North Fishtail Bay Area and the Maple River Area (l.c. Map 1). From these areas, especially the first two, we obtained the snails that furnished the cercariae for infecting the definitive hosts.

When collections of *P. parkeri* were brought into the laboratory, the snails were placed immediately in half-pint milk bottles with water and examined during the same day and also the next morning to see if cercariae of *T. physellae* had escaped from any of them. The positives were kept in the same milk bottles, but were given lettuce to eat, and the water on them was changed frequently.

Enough egg masses to produce all the laboratory raised juveniles needed were obtained from the snails examined for the cercariae. The eggs hatched in about two weeks, the time varying with the temperature. The juveniles were kept in jars and aquaria of various sizes with a few water plants, a constant supply of lettuce and a little chalk. Except in a few cases they lived fairly well but appeared to be retarded in growth.

At first, canaries were used as experimental hosts. They were kept in wire rat cages fitted with perches. For exposure to infection they were placed in small glass jars in water containing numerous cercariae from several snails. Just enough water was put in the jars to keep the birds constantly wet while permitting them to keep their footing. They were kept in the water for about one-half hour for

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each exposure. The 12 used in the summer of 1950 were given 3 to 11 exposures over periods varying from about 8 to 20 days. About 2.5 weeks after the first exposure the birds were examined to see if they were producing eggs. All examinations were made by using the type of side-arm flask designed by McMullen and Beaver (1945, Fig. 1) for isolating miracidia. This proved to be effective since, if eggs were present, the miracidia began to come up into the arms of the flasks exposed to light in about half an hour. Three of these canaries were found to be positive by this method. In spite of the large numbers of cercariae to which they had been exposed, the infections were rather light and furnished enough miracidia to infect snails for a period of only 9 days. In the summer of 1951 10 canaries were used. Only 3 became positive; 2 died soon after the eggs appeared in their droppings and the other had only a light infection. In the canaries the infections matured in about 3 weeks after the first exposure to cercariae. Fortunately, 3 domestic ducks that were also exposed to cercariae during the summer furnished miracidia for infecting snails.

Since during these 2 summers it had not been possible to make an adequate study of all stages of development of the mother and daughter sporocysts of *T. physellae* the work was continued during the summer of 1952. In the light of the experience of the previous summer only domestic ducklings were used as experimental definitive hosts. Therefore 6 ducklings about 2 weeks old were obtained. For each exposure to cercariae the ducklings were placed in covered pails for about one-half hour in enough water to cover about half of their bodies. Isolation cages with removable pans were available so that they could be separated to obtain individual fecal specimens for examination. Care was taken not to put too large quantities of feces in the flasks for isolation of miracidia for fear that the eggs would not hatch. The ducks were exposed to infection from June 21 to July 9, the number of exposures varying from 8 to 16. Beginning June 24 large numbers of pooled cercariae from 8 to 10 infected snails were used. Daily examinations were made from July 10 to July 30 and 3 more between July 30 and August 8. All 6 ducks were negative. During the latter part of this period the steps in the handling of the feces in the flasks were carefully checked and varied to make sure that positives were not being missed.

It occurred to us that the failure to get eggs in the feces of the ducks during the summer of 1952 might have been due to the development of an immunity. Therefore in the summer of 1953 we gave fewer exposures to somewhat smaller numbers of cercariae. Eight ducks were given pooled cercariae from 7 to 10 snails over a period of a few days: two were given 1 exposure; one, 2; two, 3; two, 4; and one, 6. All became positive in about 3 weeks after the first exposure.

For the experimental infections of the snails the miracidia were removed from the arms of the flasks as soon as possible after their appearance. Usually a single snail was exposed to 4 miracidia in a small stender dish, but with larger snails 8 were used. All snails for the experiments up to the last half of the summer of 1953 were raised from eggs in the laboratory. Most of them were less than 4 mm. in length when examined, and some were considerably smaller. Such small juveniles usually died in the later stages of the development of the infections. Therefore a collection of juveniles of *P. parkeri* was made from South Fishtail Bay near the Station shortly after the middle of July, 1953. They were much larger than the laboratory raised

juveniles and continued to grow well in the aquaria. They were used chiefly to study the later stages of the development of the daughter sporocysts in the digestive gland of the snail. For direct comparison with them, a large group of small laboratory raised juveniles was exposed to miracidia at the same time. The examination of these 2 groups showed that the last stages of development of the daughter sporocysts were considerably modified in the small snails so that the description of these stages will be taken from the examination of the larger juveniles. In these 2 lots the shell size was measured at the time of examination. A total of 68 large juveniles was examined from 11 to 39 days after exposure to the miracidia. Of these, 23 were infected with *T. physellae*. There was no evidence that any of these infections had been naturally acquired since they were all in the expected stage of development. Seven of the other snails harbored mature infections of other trematodes, viz., 3 monostomes, 3 echinostomes, and 1 strigeid.

The methods used in the examination of snails and in the study of the stages of sporocyst development were the same as those used in recent studies (Cort, Ameel and Van der Woude, 1953).

STRUCTURE AND DEVELOPMENT OF MOTHER SPOROCYSTS

During development of the miracidium of *T. physellae* the germinal cells have already undergone considerable multiplication. They are arranged in the body cavity of a fully developed miracidium in an elongate compact group extending from the posterior margin of the central nerve mass almost to the posterior end of the body (Ameel, Van der Woude and Cort, 1953). The number counted in 8 miracidia varied from 20 to 30 with an average of 26.

Most mother sporocysts of *T. physellae* were found in the mantle of the snail host; a very few, however, were located in the mesenteries, digestive gland or other organs. When the snails were teased apart, very small mother sporocysts were frequently found free in the dishes. Longer ones were sewed tightly into the tissues of the mantle, and those over 2 mm. long could only with difficulty be teased out unbroken. The development of the germinal material in the mother sporocysts can best be considered in relation to size since there was a considerable variation in their rate of growth, especially from the third to the eighth day after infection. For example, in 1 series of snails examined 84 to 96 hours after exposure, the length of the mother sporocysts varied from 0.1 mm. to 0.5 mm., and the number of germinal elements and the extent of their development into embryos was correlated with the differences in size. This variation became somewhat less as the mother sporocysts approached maturity, but there was also considerable variation both in the size and number of embryos produced in full grown sporocysts.

The youngest mother sporocysts studied were about 44 hours old. Two of the smallest measured 0.11 by 0.04 mm., and 0.14 by 0.06 mm. respectively. They were crowded with germinal cells, and no embryos were present. The one shown in Figure 1 contained about 70 germinal cells. A mother sporocyst was located in sectioned material of a snail 40 to 44 hours after exposure. It had a size of about 0.15 by 0.06 mm. and contained about 85 germinal cells and a few embryos in early cleavage stages. Figure 2 shows a longitudinal section of this sporocyst in which the arrangement and structure of the germinal cells can be seen more clearly than in living specimens. Practically all of them are elongate with cytoplasmic projections and a large proportion are attached to the wall. Study of the sections of this

mother sporocyst gave the impression that the germinal cells were not crowded in the body cavity and that their cystoplasmic projections formed a network connecting them with each other and with the inside of the body wall.

Five mother sporocysts were obtained from infections about 3 days old. The smallest, broken at the posterior end and considerably contracted, had a length of slightly over 0.1 mm. and a width of about 0.05 mm. It contained about 50 germinal elements of which a few were 2- and 4-cell embryos. Another of this series, somewhat contracted, measured about 0.15 by 0.06 mm. and contained about 100 germinal elements of which almost one-fifth were 2- to 4-cell embryos. The mother sporocyst of this age shown in Figure 3 measured 0.26 by 0.05 mm.; while most of its germinal material consisted of germinal cells, several 2- to 4-celled embryos were also present. The germinal elements in its body cavity were not crowded, and some granular material was present in the spaces between them. The mother sporocyst shown in Figure 4, also from the 3-day old series, was more advanced in development. It measured 0.35 by 0.05 mm. and contained more than 100 germinal elements about one-third of which were 2- to 4-celled embryos. Its wall was quite thick; the germinal material was not crowded and a number of its germinal cells appeared as if attached tightly to the inside of the wall. The largest and most advanced in development of the 3-day old mother sporocysts measured 0.34 by 0.07 mm. It contained almost 200 germinal elements, about one-half of which were small embryos. While most of them were in the 2- to 4-cell stage, a few were small "germ balls," the largest of which measured 0.25 by 0.18 mm. A large number of the germinal cells were attached tightly to the wall, and most of the embryos were located toward the center of the lumen.

As already noted, some of the mother sporocysts grew very rapidly in the first few days after infection while others lagged. The largest, found in a series of 3.5 to 4 day old infections, were slightly over 0.5 mm. in length. The most striking advance in development was an increase in the size and number of the embryos, a few of which had begun to elongate with lengths up to 0.04 mm. Most of them, however were still in the round "germ ball" stage, and a number still had 2 to 4 cells. At this stage numerous germinal cells were still present, a large proportion of which were attached to the inner surface of the body wall.

In mother sporocysts about 1 mm. in length the body cavity was usually crowded with embryos, a few of which had reached almost 0.1 mm. in length and were distinctly elongate. Most of the embryos, however, were still in the "germ ball" and cleavage stages. A number of germinal cells were present attached to the wall of the body cavity. Figure 5 depicts a mother sporocyst from a 6 day old infection, which measured 0.93 by 0.11 mm. It had a rather thick wall and was contracted at certain places along its length. A few germinal cells are shown in the drawing attached to the wall near the anterior end, and undoubtedly, some were present along its whole length.[2] The largest embryos were somewhat elongate and almost 0.1 mm. long.

[2] In mother sporocysts at this and later stages it was frequently very difficult to see the germinal cells in living specimens on account of the crowding of the embryos and the thickness of the wall. Sometimes they could be seen best in contracted regions but only by careful examination with the high power magnification. Occasionally in sporocysts in which the embryos were less crowded they could be seen much more easily. However, we were never able to make counts of the number of germinal cells with any degree of accuracy in mother sporocysts of greater length than about 0.4 mm.

The stages of development of mother sporocysts between that shown in Figure 5 and those containing daughter sporocysts about ready to escape were not studied in detail. Many of them had constricted regions separating expanded portions in which numbers of embryos of varying sizes were crowded together in large groups. In others, there were no constricted places and the whole length of the body cavity was crowded with embryos; in some cases the embryos were not so crowded. Groups of embryos frequently adhered so tightly to each other that they moved as units with the movements of the sporocyst. There was also a considerable variation in the size of the mother sporocysts at this stage and the numbers of embryos they contained. With careful examination it was usually possible to find a few germinal cells attached to the wall.

Several mother sporocysts at about the stage that daughter sporocysts were beginning to escape were studied in detail. They also varied considerably in size and in number of germinal elements. One measuring only about 1 mm. in length contained 2 daughter sporocysts about 0.30 mm. long that were moving actively. Its body cavity was not crowded and contained a number of embryos of various sizes that were not attached in groups; a few very small embryos were seen attached to the wall. A much larger mother sporocyst, at about the same stage of development, was teased out unbroken from a very small snail with a shell diameter of only about 1 mm., which had been exposed to infection 11 days previously. It had no completely contracted regions and measured about 3.5 mm. in length and from 0.05 to 0.18 mm. in width. A careful examination of the digestive gland of this snail showed no daughter sporocysts and none was found outside the mother in the migrating stage. However, in this mother sporocyst there were a few daughter sporocysts from 0.28 to 0.36 mm. in length that showed active movement. In all, 120 embryos were counted, the smallest of which were round or slightly oval with a diameter of about 0.05 mm. Forty were distinctly elongate and had a length of over 0.15 mm. In addition, there were a few smaller embryos free in the body cavity, and some very small embryos and a few germinal cells attached to the wall of the body cavity.

Two mother sporocysts from 11 day old infections at a stage slightly further advanced were found in the same series of small juveniles as the one just described. One was from a snail with a shell diameter of about 1.5 mm. Eight daughter sporocysts which varied from 0.29 to 0.33 mm. in length were found in the digestive gland of this snail. This mother sporocyst, which had no completely constricted places, had a length of about 3.7 mm. and a maximum width of 0.19 mm. The number of daughter sporocyst embryos counted in it was only 57. They were of a variety of sizes, but 46 of them were elongate and varied in length from 0.06 to 0.29 mm. The largest were active and appeared to be about ready to escape. A few scattered germinal cells could be seen attached to the wall. The other mother sporocyst which was broken when removed from the mantle of a snail with a shell size of 3 by 2.5 mm. had a total length estimated at about 4.5 mm. Twenty daughter sporocysts were found in the digestive gland of this snail. Fifty-two elongate daughter sporocysts were counted in the pieces of the mother sporocyst, the largest being 0.25 to 0.30 in length. There were about 28 long free daughter sporocysts, most of which had probably been broken out from the mother, although a few

might have represented migrating stages. A few germinal cells and small embryos could be seen attached to the wall when the pieces of this mother were examined under high magnification.[3]

The largest mother sporocyst, isolated intact, was about 5.6 mm. long. It was from an 11 day old infection in a large juvenile snail with a shell size of 13 by 8 mm., from the group that had been collected from the lake. Seventy-six daughter sporocysts, the largest of which were 0.5 to 0.6 mm. in length, were found in the digestive gland of this snail. This figure is probably much too small, since it is very difficult to count the sporocysts in the digestive gland of a snail this large. The body cavity of this mother sporocyst contained 55 elongate daughter sporocyst embryos, the largest of which measured 0.38 by 0.05 mm.; some were actively moving and appeared to be about ready to escape. In addition, about 100 round or oval embryos of varying sizes were counted. There were also large numbers of single germinal cells and 2- to 12-celled embryos attached to the wall of the body cavity along its whole length. These were, in many cases, attached to each other in compact groups, some of which were composed only of embryos while others consisted of both germinal cells and small embryos.

A 15-day old infection was studied from a large juvenile snail with a shell size of 10 by 7 mm. from the group collected from the lake. The digestive gland contained numerous daughter sporocysts, the largest of which measured 1.5 mm. long by 0.07 wide. The single mother sporocyst that was found was broken into several pieces. Some parts were completely collapsed, but others contained some well developed elongate daughter sporocysts and a few small embryos and germinal cells attached to the wall. One portion almost 1 mm. in length contained 6 large motile daughter sporocysts, 1 small embryo under 0.02 mm. in diameter, and a few germinal cells.

In some mother sporocysts still containing elongate daughter sporocysts that showed movement and appeared normal, no germinal cells could be seen attached to the wall. It was difficult to be certain of this in the examination of living material because of the difficulty of seeing the germinal cells. However, this point was checked in sections of mother sporocysts. In sections of a 16 day old infection most of the mother sporocyst was completely collapsed and devoid of embryos and germinal cells. Figure 6 is a section through a small portion of this mother sporocyst showing 2 of the few embryos that were present. They appear to be perfectly normal, but the sporocyst wall is entirely without attached germinal cells or small embryos.

The length of time required for the living germinal material in the mother sporocysts to become completely exhausted varies considerably. However all mother sporocysts found in infections more than 20 days old were much shriveled and very dark and granular. The few elongate daughter sporocysts and smaller embryos occasionally found in them were in every case immobile and appeared to be dead. In a few infections the approximate sizes of these old mother sporocysts could be determined by measuring the pieces. In 2 instances they were about 6 mm. in length.

[3] All the mother sporocysts so far described came from small laboratory raised juveniles.

STRUCTURE AND DEVELOPMENT OF DAUGHTER SPOROCYSTS

The earliest stages of the daughter sporocyst embryos of *T. physellae* resembled those described for related species (Cort, Ameel and Van der Woude, 1953, Figs. 1, 2, 3, 7 and 8, and Cort, Ameel and Van der Woude, 1954b, Figs. 13, 14 and 15).

The smallest daughter sporocyst embryo studied in detail, measured 0.12 by 0.04 mm. Thirty-five germinal elements were counted in its body cavity, 5 of which were small embryos. The body cavity was not crowded and some of the germinal cells were elongate with cytoplasmic processes extending from their ends. The wall was quite thick and some of the germinal cells were closely attached to it. The anterior plug of body-wall cells, which is characteristic of schistosome and spirorchiid daughter sporocyst embryos, was about one-sixth of the total length. The elongate daughter sporocyst embryo, a longitudinal section of which is shown in Figure 6, is at about the same stage as the one just described. A few germinal cells were in division stages and a few embryos were present, 1 of which in the 3- or 4-cell stage can be seen in the drawing. In slightly larger daughter sporocyst embryos there were larger numbers of germinal cells and a larger proportion of embryos. The one shown in Figure 7, which measured 0.12 by 0.07 mm., contained about 40 germinal cells and 10 small embryos, the largest of which was slightly beyond the 4-cell stage. The anterior plug of body-wall cells was unusually prominent in this specimen. Figure 8 depicts a daughter sporocyst embryo of about the same stage that was more extended, measuring 0.18 by 0.04 mm. Its wall was quite thin, and the anterior plug of body-wall cells was very small. Fifty germinal elements were counted in its body cavity, 8 of which were 2- to 4-celled embryos. At this stage the germinal material was not at all crowded in the body cavity, leaving spaces containing dark refractile granular material. A somewhat more advanced stage of development is shown in Figure 9. In this daughter sporocyst embryo, which measured 0.23 by 0.04 mm., the total number of germinal elements was about 80. Almost one-third of them were small embryos and few, if any, were beyond the 4-cell stage. The germinal material was not crowded in the body cavity and most of the germinal cells were in close contact with the body wall, appearing to be attached to it.

The structure of the migrating stage is illustrated by the daughter sporocyst shown in Figure 10 which measured 0.38 by 0.03 mm. It was 1 of the 20 found in the digestive gland in one of the 11 day old infections described above. It was very much extended with a rather thick wall. Sixty-five embryos were counted in its body cavity and about the same number of germinal cells are shown in the drawing, most of which are attached to the body wall. It is probable that more germinal cells were present. The body cavity was not very crowded with embryos since spaces containing granules could be seen between the germinal elements. A few of the embryos were beyond the 4-cell stage, although the largest was still very small. A daughter sporocyst somewhat more advanced in development is shown in Figure 11; it measured 0.48 by 0.06 mm. More than 60 embryos were counted in it, some of which were much larger than the others and were beginning to elongate; a few germinal cells could be seen along the wall. Figure 12 illustrates a later stage. It was from the 15 day old infection described above from a large juvenile snail. It measured 1.10 mm. in length and 0.08 mm. at greatest width.

A few of its embryos were much larger than the others and somewhat elongate. Germinal cells were probably more numerous than shown in the drawing since they were very difficult to see. In parts of the body closely packed groups of embryos were separated by constrictions of the body wall. A section through the anterior half of a daughter sporocyst of about the same stage of development as Figure 12 is shown in Figure 13. It shows one elongate embryo larger than the others, but most of them are round or oval and not more than 0.035 to 0.040 mm. in diameter. Germinal cells and small groups of germinal elements are attached to the wall. The large number of germinal cells attached in the anterior tip is characteristic, and it is in this region that they persist longest. The sporocyst from which this drawing was made was unusual in the amount of empty space in the anterior part. Some daughter sporocysts at about the stage shown in Figure 12 had their embryos separated into series of densely packed groups separated by constrictions of the body wall. In such groups there were a few embryos much larger than the others and smaller ones of all sizes held tightly together. One such group shown in Figure 14 had more than 20 embryos. Some daughter sporocysts with this type of arrangement of germinal material contained from 100 to almost 200 embryos. Usually a few germinal cells could be seen in them either attached to the embryo groups or to the wall.

It is characteristic of the stage of daughter sporocyst development in which the largest cercarial embryos begin to show division into body and tail, that a few of these embryos have grown much larger than the others. This is even more striking in large almost full grown sporocysts containing large cercarial embryos with eye-spots, and in those in which the first cercariae have reached maturity. These stages will be described from the series of infections in the large juvenile snails collected from the lake.

In an infection about 20 days old in a snail measuring 13 by 9 mm., 2 mother sporocysts were found that were very dark and shrivelled and appeared to be dead. All parts of the digestive gland contained large numbers of daughter sporocysts which appeared to be of 2 to 3 mm. in length, although it was almost impossible to free any of them unbroken for accurate measurements. They were not crowded and were for the most part located between the lobules of the gland, which did not appear to be abnormal. This snail, like almost all the series of large juveniles in which there were mature or almost mature infections, had been active and appeared to be in good condition when examined. Only a few smaller daughter sporocysts appeared to be developing normally. There were also numbers of small dead sporocysts from about 0.15 to 0.60 mm. in length in various stages of degeneration. No mature cercariae were found in this infection, although in another infection of the same age in this series a few were present.[4]

Two infections in large juveniles from which cercariae had been escaping for 5 days were examined on about the 29th and 30th days after exposure. All the living daughter sporocysts in them were large and were crowded with cercarial embryos with eye-spots and some mature cercariae. When the shells of these

[4] It was not difficult to tell mature cercariae from large almost mature embryos. Those not quite mature swam slowly and awkwardly when broken out of the sporocysts and their two anterior pairs of eosinophilic glands were dark and granular. Mature forms swam actively and their four anterior glands were hardly darker or more granular than the posterior 3 pairs of basophilic glands.

snails were removed, large numbers of actively swimming mature cercariae were freed. In one snail no small dead sporocysts were found, and in the other there was a considerable number.

An infection was examined about 33 days after exposure in a snail measuring 13 by 8 mm. from which cercariae had been escaping for 9 days. In it in addition to many sporocysts crowded with large cercariae there were a few that appeared perfectly normal, but were collapsed for most or all of their length. Some contained 1 or 2 large cercarial embryos, and others none. When freed from the snail's tissue, these almost empty sporocysts moved actively. Infections examined on the 35th, 37th, and 39th days from which cercariae had been escaping for 11, 13, and 15 days respectively had larger numbers of collapsed, almost empty sporocysts. These infections also still contained numerous sporocysts crowded with large cercarial embryos. No older infections were available in this series, but it is evident that those that had given off cercariae for 9 to 15 days would have continued to shed them for a longer period since numbers of large almost mature cercarial embryos were still present.

Perhaps the most striking thing in these mature infections was that all the living daughter sporocysts were of large size and contained well-developed embryos. Mature cercariae of this species have a total length of almost 1 mm. and the embryos with eye-spots are almost as large. It is evident, therefore, that only a few embryos can develop simultaneously to large size in sporocysts about 3 mm. in length. In fact, the small numbers of almost mature cercariae present in such sporocysts crowded their body cavities. When the shells were removed from the snails harboring mature infections, the anterior ends of numerous sporocysts were seen protruding from the surface of the digestive gland, and cercariae could be seen escaping from their birth pores. In daughter sporocysts at this stage there were usually only a few embryos of intermediate sizes between small round germ balls and embryos with eye-spots. The remainder of the germinal material consisted of a few small single embryos, free groups of embryos some of which contained germinal cells, and sometimes a few germinal cells or small groups of germinal material attached to the wall. A few examples of individual sporocysts will help to make this clear. One about 3 mm. in length contained 18 large cercarial embryos with eye-spots, a few single embryos of intermediate stages and a number of small groups of germinal elements. This was the greatest number of large cercarial embryos seen in a daughter sporocyst. Another sporocyst measuring 2.4 mm. by 0.06 to 0.13 mm. had 11 large cercarial embryos which crowded its body cavity and constantly changed its shape by their movement. A few single embryos and groups of germinal elements were also present.

In the infections of this series from which cercariae had been escaping for 9 to 15 days some of the daughter sporocysts appeared almost empty and contracted. Such sporocysts might contain a few small single embryos, but most of their germinal material consisted of groups of germinal elements. In most cases no germinal elements could be found attached to the wall, but, occasionally, a few small embryos or even germinal cells were present at the anterior end. One daughter sporocyst about 2.5 mm. long contained 1 large active cercarial embryo and nothing else except about 15 small groups of germinal elements. These sporocysts were almost completely collapsed, but they were evidently not degenerating and showed

movement when separated from the snails' tissue. The small separate embryos and the groups of germinal elements contained by these sporocysts moved freely in the body cavity and sometimes were seen collected together in slightly inflated areas especially at the anterior and posterior ends (Fig. 15).

A careful study was made of these groups of germinal elements from a number of large daughter sporocysts. Some groups consisted of a few embryos which in some cases appeared about ready to break apart; others were composed of rather loosely attached embryos and a few germinal cells. A common type were rather discrete structures usually containing a few embryos and one or two germinal cells. Sometimes these groups were elongate with the germinal elements in linear arrangement. In a few cases such groups were attached to the wall. In a few groups a single germinal cell was attached to 1 or 2 small embryos, and a few were composed of 3 or 4 germinal cells.

MODIFICATION OF INFECTIONS PRODUCED BY DEVELOPMENT IN SMALL SNAILS

The most detailed information on the effect of small snails on the development of experimental infections of *T. physellae* came from the examination of laboratory raised juveniles infected on the 24th of July, 1953 at the same time as a group of juveniles collected from the lake. In the small snails, almost all of which were less than 4 mm. in length, the daughter sporocysts appeared to develop normally until considerable numbers of them had migrated to the digestive gland. However, in later stages, as the sporocysts began to increase rapidly in size and became more and more crowded, the whole character of the infections was greatly modified. Only a small number of the daughter sporocysts grew normally and produced large cercarial embryos. The others died while still small and were found in various stages of disintegration all through the digestive gland. It seemed as if the first daughter sporocysts that migrated into the digestive gland had preempted the available space, creating conditions unfavorable for the development of those that arrived later. Most of the snails died before the first cercariae matured, and very few survived for more than a few days after the first began to escape.

A few examples will illustrate the extent to which the infections were modified in the small juveniles. In a snail with a shell size of 3 by 2 mm. with an infection about 25 days old, there were only 12 large living daughter sporocysts which contained a few large cercarial embryos with eye-spots. All the other daughter sporocysts were dead. Of 2 snails, measuring 3 by 2 mm., and 5 by 3 mm., respectively, examined 27 days after infection, each had about 25 large normal daughter sporocysts containing a few large cercarial embryos with eye-spots. Numerous dead sporocysts of various sizes were present all through their digestive glands. Neither of them had given off cercariae although they had been isolated in bottles for 4 days before examination. A few apparently mature cercariae were found in the larger one. In a 28 day old infection in a snail 3 by 2.5 mm. there were only 6 large normal sporocysts in the digestive gland. No cercariae had escaped for 5 days before examination and only 3 of the small number of large cercarial embryos appeared mature. In two infections in snails about 3 mm. in length a few cercariae escaped on the 28th day after infection. Another infection in a snail of about the same size examined on the 29th day was much more retarded. There were about 30 large daughter sporocysts in its digestive gland. None of the cercarial embryos had eye-spots

and only a few showed division into body and tail. Records of experimental infections in small snails examined during 1950 and 1951 showed similar modifications. Ten examined 17 to 19 days after infection contained only small numbers of large living daughter sporocysts. Only a few of the largest cercarial embryos showed division into body and tail. In some infections 22 to 23 days old none of the largest cercarial embryos had eye-spots. The earliest in which there were mature cercariae was 27 days old. The digestive gland of this snail contained a few daughter sporocysts almost 3 mm. in length containing large cercarial embryos with eye-spots, a few of which appeared to be almost ready to escape.

It seems, therefore, that in small snails 3 mm. in length or less the development to maturity of infection of *T. physellae* is delayed for about a week. In them only a comparatively few daughter sporocysts live and develop normally, which would greatly reduce the number of cercariae produced. In such infections large numbers of dead sporocysts in different stages of development are found in the digestive gland. Under the conditions of the experimental infections in the laboratory, almost all the small snails died just before or just after the first cercariae matured, while almost all the larger snails lived well, some producing cercariae 5 to 15 days before examination.

SUMMARY AND CONCLUSIONS

During the development of the miracidium of *T. physellae* there is already a considerable multiplication of the germinal cells. This continues in the very young mother sporocysts both before and after the first embryos appear. More and more embryos are produced as the mother sporocysts grow and the rapid multiplication of germinal cells continues. About the time the first daughter sporocyst embryos develop beyond the 4- to 8-cell stage, the early phase of rapid multiplication of germinal cells has been completed and most of those remaining are closely attached to the wall. Most of the embryos grow slowly and are still quite small at the time the first daughter sporocyst escapes from the mother sporocyst at about 10 days after infection. In the next 5 or 6 days the growth is very rapid and the migration is practically completed; soon after that the mother sporocyst dies. The daughter sporocysts grow rapidly in the digestive gland of the snail and by about 21 to 24 days after infection, when the first cercariae begin to mature, all appear to have reached their full size and almost all of them contain a few cercarial embryos with eye-spots. Any smaller ones present are dead and disintegrating. Fully developed daughter sporocysts from which no cercariae have escaped usually contain about 10 to 20 large cercarial embryos with well developed tails and eye-spots which on account of their large size crowd the body cavity.

In infections in larger juveniles from which cercariae have been escaping for 9 to 15 days, numbers of partially collapsed daughter sporocysts are found from which most or all of the large cercariae have escaped. In them only a few cercarial embryos in intermediate stages of development are ever found. Such daughter sporocysts appear to be alive and normal and move actively when freed from the tissues of the snail. They contain a reserve of germinal material consisting of small groups of germinal elements which float freely in the body cavity, many of which contain germinal cells. Part of these groups in which the embryos are rather loosely attached to each other appear to be derived from the breaking up of larger

groups by the activity of the cercariae. Other groups which are more compact have probably developed late from germinal cells attached to the wall. In fact, similiar groups are sometimes seen attached to the wall. Sometimes, also, a few single germinal cells and small groups of germinal material can be found attached to the wall of the body cavity especially near the anterior end. These partly empty daughter sporocysts were seen only in our series of large infected juveniles during the last half of August. There seemed to be enough large cercarial embryos left in these infections to keep up the production of cercariae for a few weeks more. Considering how cold the weather becomes in September and October in the Douglas Lake region, it seems doubtful that the very small embryos and especially any of the germinal cells would produce mature cercariae until the next spring. It seems probable, also, that some of the remaining germinal cells would continue to divide. Therefore it can be suggested that in juveniles infected about the middle of July or even earlier the infections would not be exhausted when their development was stopped by cold weather, but would produce considerable numbers of mature cercariae early the next summer.

In small laboratory raised juveniles of *P. parkeri* the infections are greatly modified in the later stages of development. Most of the smallest snails die before or just after their infections reach maturity. Death seems to be caused by injury to the digestive gland produced by the rapid increase in size of the daughter sporocysts during the final stages of growth. About a week longer is required for the infections to mature in the small snails than in the larger ones. Also, only a small proportion of the daughter sporocysts that migrate into the digestive gland grow to full size and produce cercariae while others die soon after migration. These findings suggest that in the small snails the development of the daughter sporocysts under crowded conditions produces some modification in the physiology of the host that limits the numbers that develop and causes the death of the others. Sometimes dead daughter sporocysts are also found in the larger juveniles especially when more than one mother sporocyst is present.

It seems probable that the course of development is much the same in natural as in experimental infections. Any snails infected soon after hatching would probably be killed by the destruction of the digestive gland, and slightly larger ones, if they survived, would produce only a few cercariae. It seems probable, also, that snails infected when they are almost full grown would produce larger numbers of cercariae than even the largest juveniles in our experimental infections, because larger numbers of daughter sporocysts would find conditions favorable for their development in the much larger digestive glands. In mature infections of *T. physellae* in adult snails examined in the early summer, the digestive glands which are many times larger than in even the largest juveniles in our series are filled with daughter sporocysts and give off cercariae in large numbers. Evidently, therefore, the course of development of infections of *T. physellae* is modified by the time of the year at which the miracidia penetrate. Since development takes only a little over 3 weeks, it is possible that a few of the infections in juveniles which hatched early in the season would run their course and be almost exhausted before winter. Those infected about the middle of the summer would give off numerous cercariae both before and after the winter rest period. Any infected after late August would probably not produce cercariae until the next spring, and those in-

fected as adults in the early spring would have a still more limited period of cercarial production. Since the adults of *P. parkeri* begin to die off soon after the end of the breeding season, many infections would be terminated by the death of their hosts before completion of their production of cercariae. The study by Cort, Olivier and McMullen (1941) on natural infections of *T. physellae* gives support to these suggestions.

The mother sporocysts of *T. physellae* resemble closely those in experimental infections of the spirorchiid, *C. elephantis*, and a closely related species, *T. stagnicolae* (Cort, Ameel and Van der Woude, 1954b and c). They differ greatly from the large inflated mother sporocysts of *Schistosomatium douthitti* (Cort, Ameel and Olivier, 1944). They also differ from the mother sporocysts of *T. stagnicolae* that were found in naturally infected adult snails (Cort and Olivier, 1943). The daughter sporocysts of *T. physellae* in their early stages of development contain more germinal elements than those of *C. elephantis*. They are also quite different in the arrangement of the germinal material from the larger daughter sporocysts of *T. stagnicolae* found in natural infections of adult snails, but are similar to those of this species in experimental infections in laboratory raised juveniles. In the rapidity of development in the intermediate host, *T. physellae* resembles *T. stagnicolae*, while infections of *C. elephantis* and *S. douthitti* require more time to develop to maturity.

In all these species the germinal cells are scattered along the whole length of the sporocysts and are usually attached to the wall, and no germinal masses that serve as centers of multiplication like those found in certain other groups of the digenetic trematodes are present.

References Cited

Ameel, D. J., Van der Woude, Anne, and Cort, W. W. 1953 Studies on the miracidium of the genus Trichobilharzia with special reference to the germinal cells. Proc. Helminth. Soc. Washington **20**: 40–42.

Cort, W. W. 1950 Studies on schistosome dermatitis XI. Status of knowledge after more than twenty years. Am. J. Hyg. **52**: 251–307.

———, Ameel, D. J. and Olivier, Louis 1944 An experimental study of the development of *Schistosomatium douthitti* (Cort, 1914) in its intermediate host. J. Parasit. **30**: 1–17.

———, ——— and Van der Woude, Anne 1953 Further studies on the early development of the daughter sporocysts of *Schistosomatium douthitti*. Proc. Helminth. Soc. Washington **20**: 43–49.

———, ——— and ———1954a Germinal development in the sporocysts and rediae of the digenetic trematodes. Exp. Parasitol. **3**: 185–225.

———, ——— and ———1954b Germinal development of the turtle blood flukes. Proc. Helminth. Soc. Washington **21**: 85–96.

———, ——— and ———1954c Further studies on the germinal development in the sporocysts of a bird schistosome, *Trichobilharzia stagnicolae* (Talbot, 1936). Proc. Helminth. Soc. Washington **21**: 97–106.

———, Olivier, Louis and McMullen, D. B. 1941 Larval trematode infection in juveniles and adults of *Physa parkeri* Currier. J. Parasit. **27**: 123–141.

——— and ———1943 The development of the sporocysts of a schistosome, *Cercaria stagnicolae*, Talbot, 1936. J. Parasit. **29**: 164–176.

McLeod, J. A. and Little, G. E. 1942 Continued studies on cercarial dermatitis and the trematode family Schistosomatidae in Manitoba. Canadian J. Res. **20** (Sect. D): 170–181.

McMullen, D. B. and Beaver, P. C. 1945 Studies on schistosome dermatitis. IX. The life cycles of three dermatitis producing schistosomes from birds and a discussion of the subfamily Bilharziellinae (Trematoda: Schistosomatidae). Am. J. Hyg. **42**: 128–154.

TALBOT, S. B. 1936 Studies on schistosome dermatitis. II. Morphological and life history studies on three dermatitis-producing schistosome cercariae, *C. elvae*, Miller, 1923, *C. stagnicolae* n. sp., and *C. physellae* n. sp. Am. J. Hyg. **23:** 372-384.

DESCRIPTION OF PLATES

PLATE I

Mother sporocysts of *Trichobilharzia physellae*.

FIG. 1. Very young mother sporocyst, 0.14 by 0.06 mm., 44 hours old.

FIG. 2. Section of very young mother sporocyst, 40 to 44 hours old.

FIG. 3. Very young mother sporocyst, 0.26 by 0.05 mm., about two and one-half days old.

FIG. 4. Three day old mother sporocyst, 0.35 by 0.05 mm. (Cort, Ameel and Van der Woude, 1954a, Fig. 47)

FIG. 5. Six day old mother sporocyst, 0.93 by 0.11 mm.

FIG. 6. Section of a portion of a mother sporocyst from 16 day old infection containing well developed daughters, but no small embryos or germinal cells.

PLATE II

Daughter sporocysts of *T. physellae*.

FIG. 7. Daughter sporocyst embryo, 0.12 by 0.07 mm.

FIG. 8. Daughter sporocyst embryo, 0.18 by 0.04 mm., about the same stage as Figure 7 but more extended.

FIG. 9. Daughter sporocyst embryo, 0.23 by 0.04 mm.

FIG. 10. Young daughter sporocyst from the digestive gland of host, 0.38 by 0.03 mm. (Cort, AMEEL AND VAN DER WOUDE, 1954a, Fig. 49).

FIG. 11. Young daughter sporocyst, 0.48 by 0.06 mm.

FIG. 12. Young daughter sporocyst, 1.1 by 0.08 mm.

FIG. 13. Section of anterior end of young daughter sporocyst about the same stage as Figure 12 showing germinal elements.

FIG. 14. Small portion of daughter sporocyst showing group of cercarial embryos.

FIG. 15. Anterior end of daughter sporocyst from which cercariae have escaped, showing groups of germinal elements.

PLATE I

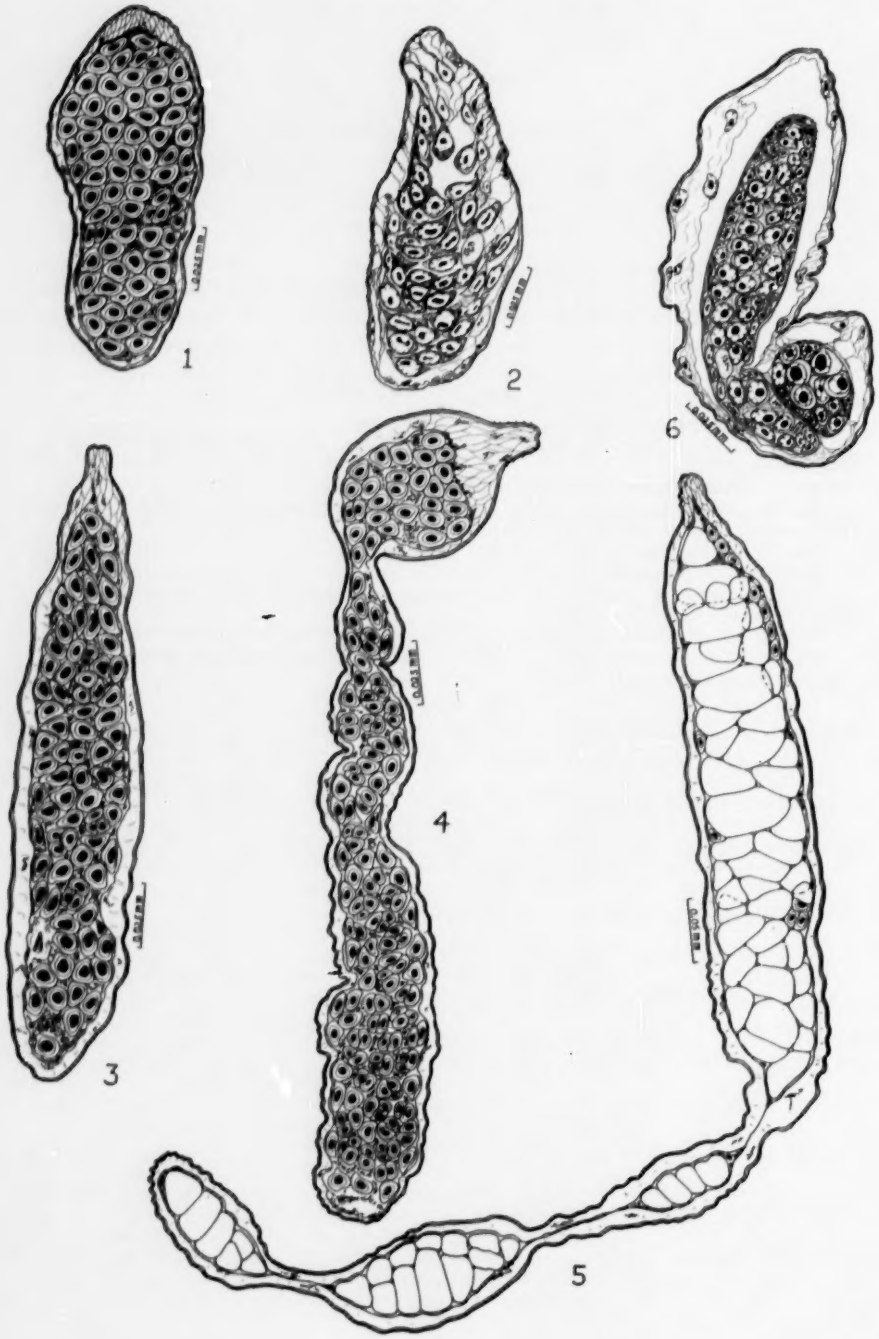

PLATE II

STUDIES ON *TRICHINELLA SPIRALIS*
I. MALE AND FEMALE REPRODUCTIVE SYSTEMS

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Trichinella spiralis is still an important parasite of man in various parts of the world and consequently is of considerable interest from many points of view. However, few fundamental studies have been made in recent years on the microscopic anatomy of the parasite itself. This paper is a redescription of the male and female reproductive systems.

MATERIALS AND METHODS

The adult worms in this study were obtained chiefly from white mice, occasionally from white rats, which were usually killed within 3 to 7 days of infection. The infected animals were left unfed, but given water, for about 24 hours before being killed for examination. The intestine was opened and kept in normal saline at body temperature while the worms were being removed. The worms were best studied while still active; within two hours after the host was killed. The physical activity of the various parts of the worm facilitated the understanding of the systems. In fact, the study was chiefly made with living materials.

Studies were also made from dissected specimens. A "00" insect pin, further sharpened on an oil stone, served as the dissecting needle. Under stereoscopic microscope a single puncture was made at the posterior end of the worm whereupon a great portion of the genital system would immediately be expelled to the exterior by pressure within the parasite.

Dissected and entire worms were also fixed and stained with haematoxylin and alum cochineal, while glycerine mounted specimens were also prepared. The study and the camera lucida drawings were made by means of a Leitz Ortholux microscope.

THE MALE REPRODUCTIVE SYSTEM

(Plate I, figs. 1–6)

The male reproductive system is a single tube. As in most nematodes, it is differentiated into four regions: testis, vas deferens, seminal vesicle and ejaculatory duct (Fig. 1). In addition, there is a copulatory tube involved in the formation of the bell-shaped copulatory structure.

Testis: The testis (Fig. 1, t) is a thick-walled tube occupying most of the posterior half of the body cavity. It begins a short distance from the cloaca, near the anterior end of the seminal vesicle and extends forward as a prominent structure. As it approaches the posterior extremity of the stichosome (the column of esophageal gland cells), its diameter is somewhat reduced and it turns abruptly backward just behind the stichosome, as if this voluminous organ prohibited its forward development. Thereafter, it lies ventrally in the body cavity. After the testis passes backward, at about one-third of the length of the forward portion, its diameter is reduced and it is continued as the thin-walled vas deferens.

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The tubular testis is closed at one end. The germinal cells, instead of being located at the distal end of the testis as in many other nematodes, are found on the wall of the testis throughout its whole length. The wall consists of several layers of large cuboidal cells (Fig. 2) which are sometimes stretched into a columnar shape. These cells have richly protoplasmic contents and large nuclei. Various stages of spermatogenesis were seen along the free border of these cells. The whole lumen is full of spherical sperm cells. The wall of the testis, as well as the other regions of this system, shows a peristaltic movement that, coupled with the body movement, creates a churning action, which assures the passage of the sperm cells.

Vas deferens: The vas deferens (Fig. 1, vd) is a thin-walled narrow tube lying ventral to or on one side of the testis. It gradually becomes dilated and is continued as the somewhat inflated seminal vesicle which is full of sperm cells. A valve between the testis and the vas deferens or between the vas deferens and the seminal vesicle, if present, could not be determined.

Seminal vesicle and ejaculatory duct: The seminal vesicle (Fig. 1, sv) is a dilated tube for the storage of sperm cells and is a very conspicuous structure at the posterior end of the body. It is partly overlapped by or posterior to the testis. The wall of its posterior end, as well as that of the ejaculatory duct, is very muscular and granular.

The seminal vesicle is continued posteriad by a narrower tube with a very muscular and granular wall and might be considered as an ejaculatory duct (Fig. 1, ej), although there is no distinct demarcation between the seminal vesicle and the ejaculatory duct. Sometimes the ejaculatory duct was seen quite dilated, but because it contracts into a fairly narrow tube while sperm cells are being pushed into the copulatory tube, it functions as an ejaculatory duct.

The intestine usually lies dorsal or lateral to the seminal vesicle. However, its posterior end always takes the dorsal position and enters the cloaca dorsally, while the ejaculatory duct takes the ventral position and enters the cloaca ventrally, immediately behind the entrance of the intestine (Figs. 3 and 4).

Cloaca and copulatory tube: The cloaca is a fairly muscular structure (Figs. 3 and 4, c), attached to the body wall by a thin membrane (Fig. 3, vs), the ventral suspending membrane, along about two-thirds of its length. This membrane is very elastic and was seen stretching out to a great distance when the cloaca was dragged forward. More often, it was seen in a contracted condition and the cloaca was closely in contact with the ventral body wall. Consequently, the cloaca has never been found free from the ventral wall, whereas a spacious body cavity was often seen between the dorsal body wall and the cloaca. At the distal end of the cloaca there are two groups of muscles (Fig. 3), each group having one end inserted into the dorsal body wall and the other attached to the cloaca. In addition, minor groups of muscles (Fig. 4) are present at the posterior extremity of the body.

Inside the cloaca there is a thin-walled tube, which the writer calls a copulatory tube (Fig. 4, ct), because it is involved in the formation of the bell-shaped copulatory structure (seen in Fig. 6, cb). The ejaculatory duct and the intestine do not discharge into the cloaca, but into the copulatory tube. From a living specimen, under pressure of the cover glass, sperm cells were seen flowing from the ejaculatory duct into the copulatory tube, and some of the sperm cells were seen flowing

forward into the intestine, indicating that both the ejaculatory duct and the intestine are continuous with the copulatory tube, although the actual connection could not be seen. Usually the copulatory tube was empty and folded. The distal portion of the copulatory tube is a short and narrower tube which opens to the exterior through the cloacal opening between the copulatory appendages (Fig. 5).

The copulatory tube is supported dorsally by a bundle of strong muscles, the copulatory muscle (Fig. 3, cm), which is about three-fifths the length of the tube. The copulatory muscle emerges from the cloacal wall at its dorsal anterior angle, at the junction of the intestine and the cloaca, where the copulatory muscle unites with the longitudinal muscles, the outer layer of muscle, from the digestive tract to form a heavy muscular bundle (Figs. 3 and 6, cm) lying in the body cavity. This copulatory muscle is attached to the dorsal body wall a short distance behind the tip of the testis.

The copulatory bell is formed by the extrusion of the copulatory tube through the cloacal opening as the result of the contraction of the cloaca and the body wall and the coincident relaxation of the copulatory muscle (Fig. 6). In a living worm sperm cells were seen flowing out through the copulatory bell.

Copulatory appendages and accessory papillae: The copulatory appendages (Figs. 3, 4, and 5) are two large ventrally bent appendages at the posterior extremity of the body. These appendages possibly serve as claspers during copulation, as suggested by previous workers. In addition, two pairs of papillae lie between the copulatory appendages, a pair each of conical papillae and of tubercle papillae. The conical papillae, one on each side, are closer to the cloacal opening, whereas the tubercle papillae are behind and external to the conical papillae. These might be sensory in function.

THE FEMALE REPRODUCTIVE SYSTEM

(Plate II, figs. 7–12)

The female reproductive system consists of a single ovary, oviduct, seminal receptacle, uterus and vagina. It is a voluminous system, practically filling the whole body cavity from the caudal end to the stichosome.

Ovary: The ovary (Fig. 7) is much shorter than the uterus. It fills the caudal end of the body cavity into which it is compactly packed. Its structure is best studied from dissected specimens. It is solid ventrally with an ovarian cavity dorsally (Figs. 7 and 8, ov). The germinal region thus lies along one side of the ovary instead of at its distal end.

The germinal region (Fig. 8) appears as a richly protoplasmic layer, from which the ova develop. Young ova appear as layers of closely packed flat cells each containing a large nucleus. They gradually increase in size and assume a more or less cuboidal form as they are pushed toward the lumen. The full sized ova eventually detach themselves and become free in the ovarian cavity. These free ova are of irregular form, usually oblong; only a few were seen at a time.

The wall of the ovary, except the germinal region, is very thin. The tip of the ovary is not free in the body cavity; it lies ventral to the rectum and appears to be attached to the wall of the rectum, for the tip of the ovary was often not expelled to the exterior even when the dissecting puncture was made close to the posterior

extremity of the body. The ovarian wall shows a peristaltic movement which forces the free ova to move forward toward the oviduct.

Oviduct: In dissected specimens, the oviduct appears as a short, undistended portion (Figs. 8, 9, and 10a, od) which discharges into the seminal receptacle at one side and from which it is suspended by a very thin membrane (Fig. 9), and against which the oviduct was often seen to lie. Under sufficient pressure of a cover-glass, the wall of the oviduct shows a strong muscular appearance (Fig. 10b). Vacuoles of various sizes were often seen in the wall near its entrance to the seminal receptacle. Cells at this region might be secretory in function. A valve between the oviduct and the seminal receptacle could not be determined. However, as sperm cells were never found in the ovary, there appears to be some mechanism which prevents their passage into the ovary.

Seminal receptacle and uterus: The seminal receptacle and the uterus form a continuous straight tube (Fig. 8, sr and u) and, morphologically, there is no constriction or any other demarcation between them. The seminal receptacle is a large blind sac with a wall comparatively thicker than that of the uterus and it is full of sperm cells.

The uterus is a dilated thin-walled tube full of developing embryos ranging from one-celled stage near the seminal receptacle to full-grown larvae near the vagina, with progressive stages in between. As the uterus approaches the stichosome its diameter is reduced to a narrow tube, the vagina. The uterus, like the ovary, shows peristaltic movement which might help to move the developing embryos toward the anterior end. However, mature larvae, free from the embryonic membranes, are able to assume active movement.

Vagina and genital opening: The vagina (Fig. 11) is a narrow tube differentiated into 2 regions: a longer thin-walled and a shorter thick-walled terminal portion. Larvae pass through one at a time. The thick-walled region appears to consist of 2 layers of tissue: an outer muscle layer and an inner layer of large cells with large nuclei. The number of these cells was not determined. The inner lining of the vagina is a continuation of the cuticle from the body surface. How far the cuticle extends inward could not be determined.

The female genital opening appears as a slightly elevated papilla (Figs. 11, 12) on the ventral side at about one-fourth of the body length from the anterior end. Two groups of muscles (Fig. 12), one on each side, join the vagino-genital opening area to the lateral aspect of the body wall. These muscles might function in regulating the action of this area.

DISCUSSION

The germinal cells in the testis of *Trichinella spiralis* are located on the entire *trichiura* as described by Eberth (1860) and that of *Trichuris suis* by Rauther (1918), extends for the entire length of the ovary on one side. However, it is a continuous zone instead of having a series of germinal areas or "pocket-like pouches" as in *T. trichiura.*

The germinal cells in the testis of *Trichinella spiralis* are located on the entire circumference of the testis. Whether the whole wall of the testis is comprised of germinal cells or the germinal cells are evenly distributed along the entire wall was not determined.

The formation of the copulatory bell has long been considered to be the result of the eversion of the cloaca. After weeks of study on the copulatory tube alone using living worms, the writer has come to the conclusion that it is the copulatory tube that forms this structure. The copulatory tube has an inconspicuous membranous wall which is free from the wall of the cloaca and is folded when empty. Its existence was best followed in some living worms in which the copulatory bell was not yet formed and when the sperm cells were being pushed into this folded empty tube, which became distended by the inflowing of sperm cells and assumed its tubular structure. Thus the copulatory tube takes over the function of a cloaca and serves as the "real cloaca" since both the digestive and the reproductive systems enter this tube. The whole cloacal structure appears to be a tube within a tube. No contents have been found in the space between the copulatory tube and the wall of the cloaca.

Hemmert-Halswick and Bugge (1934) described an eversible cloaca that had the form of a short, thick spindle, which might have been a partially collapsed copulatory bell, as it was often seen not fully distended in both living and preserved specimens.

Pagenstecher (1865) stated that there were 2 muscles, musculi retractores cloacae, connected with the action of copulation and which functioned in the withdrawal of the bell-shaped structure after copulation (Pagenstecher, 1865, and Gould, 1945). In the present study, only one muscle bundle was found. It is involved in the formation and withdrawal of the copulatory bell. Therefore, the name copulatory muscle is used, as its function is more than retractory. The cells of the ventral body wall where the ventral suspending membrane is attached, especially those near the anterior angle of the cloaca, are well developed. Under contraction, when the cloaca is in close contact with the body wall, those cells appeared as a bundle of muscle. This might have been mistaken for one of the retractor muscles.

Regarding the entrance of the reproductive system into the cloaca, it is stated that the reproductive system of all groups of nematodes except TRICHUROIDEA, enters the cloaca from the ventral side (Chitwood and Chitwood, 1950). Rauther (1918) found that the vas deferens of *Trichuris suis* enters the cloaca dorsolaterally. However, the present study has shown that the ejaculatory duct of *Trichinella spiralis*, as in the majority of nematodes, enters the cloaca ventrally, immediately behind the entrance of the intestine.

SUMMARY

The morphology of the reproductive systems of both sexes of *Trichinella spiralis* have been re-described in detail and new information has been presented.

In the male system, the testis is a hollow tube. The germinal cells are found on the wall of the testis throughout its whole length and various stages of spermatogenesis were seen along the free border. The ejaculatory tube enters the cloaca ventrally. A copulatory tube functions as a "real cloaca." The copulatory bell is formed by the extrusion of the copulatory tube through the cloacal opening as the result of the contraction of the cloaca and the body wall and the coincident relaxation of the copulatory muscle.

In the female system, the ovary is terminal and has its germinal zone on its ventral wall where ova are formed and gradually pushed toward the ovarian cavity

and set free. The seminal receptacle is a large blind pouch with the oviduct entering at its side. The vagina is a narrow tube consisting of a thin-walled proximal portion and a thick-walled distal portion with a number of large cells forming the inner wall of the latter.

References Cited

Chitwood, B. G. and Chitwood, M. B. 1950. An introduction to nematology. Section 1. Anatomy. 213 pp. Baltimore.

Eberth, C. J. 1860. Die Generationsorgane von *Trichocephalus dispar*. Ztschr. Wissensch. Zool. **10**: 383–400.

Gould, S. E. 1945. Trichinosis. 356 pp. Springfield.

Hemmert-Halswick, A. and Bugge, G. 1934. Trichinen und Trichinose. Ergebn. Allg. Path. u. Path. Anat. **28**: 313–392.

Pagenstecher, H. A. 1865. Die Trichinen. Nach Versuchen im Auftrage des Grosserzoglich Badischen Handelsministeriums ausgeführt am zoologischen Institute in Heidelberg von Christ. Jos. Fuchs und H. Alex. Pagenstecher. 116 pp., Leipzig.

Rauther, M. 1918. Mitteilungen zur Nematodenkunde. Zool. Jahrb. Abt. Anat. **40**: 441–514.

Explanation of Plates

Drawings were made with the aid of a camera lucida. Abbreviations: c, cloaca; cb, copulatory bell; cm, copulatory muscle; ct, copulatory tube; ej, ejaculatory duct; i, intestine; od, oviduct; ov, ovary; sr, seminal receptacle; sv, seminal vesicle; t, testis; u, uterus; vd, vas deferens; vs, ventral suspending membrane.

Plate I

Fig. 1. Male reproductive system expelled from a puncture made at the posterior end of worm.

Fig. 2. A portion of distal end of testis.

Fig 3. Male, posterior end with empty folded copulatory tube and with copulatory muscle lying on its dorsal aspect, right copulatory appendage not shown.

Fig. 4. Male, posterior end with copulatory tube being distended by the in-flowing of sperm cells.

Fig. 5. Male, caudal end, ventral view, showing copulatory appendages, papillae, and cloacal opening.

Fig. 6. Male, showing copulatory bell, copulatory muscle, with left copulatory appendage not shown.

Plate II

Fig. 7. Ovary and oviduct held by membrane lying against seminal receptacle.

Fig. 8. Anterior portion of ovary, oviduct, seminal receptacle and part of uterus.

Fig. 9. Oviduct shown suspended by a thin membrane.

Fig. 10a. Oviduct undistended.

Fig. 10b. The same oviduct under pressure of cover glass, showing its muscular wall.

Fig. 11. A portion of female showing genital opening, esophageal gland cells (stichocytes) and the vagina with a single larva.

Fig. 12. Female, area of genital opening.

PLATE I

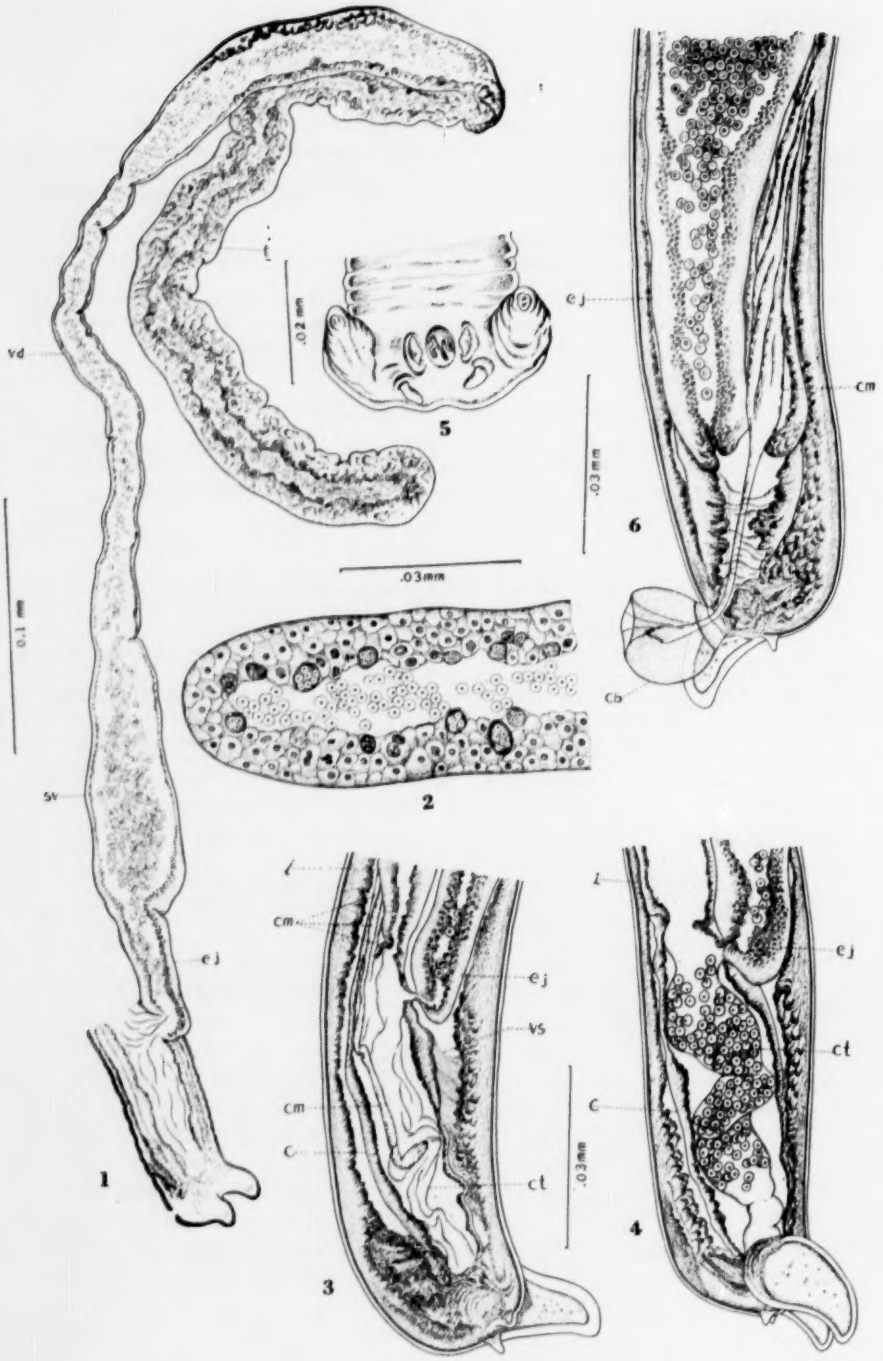
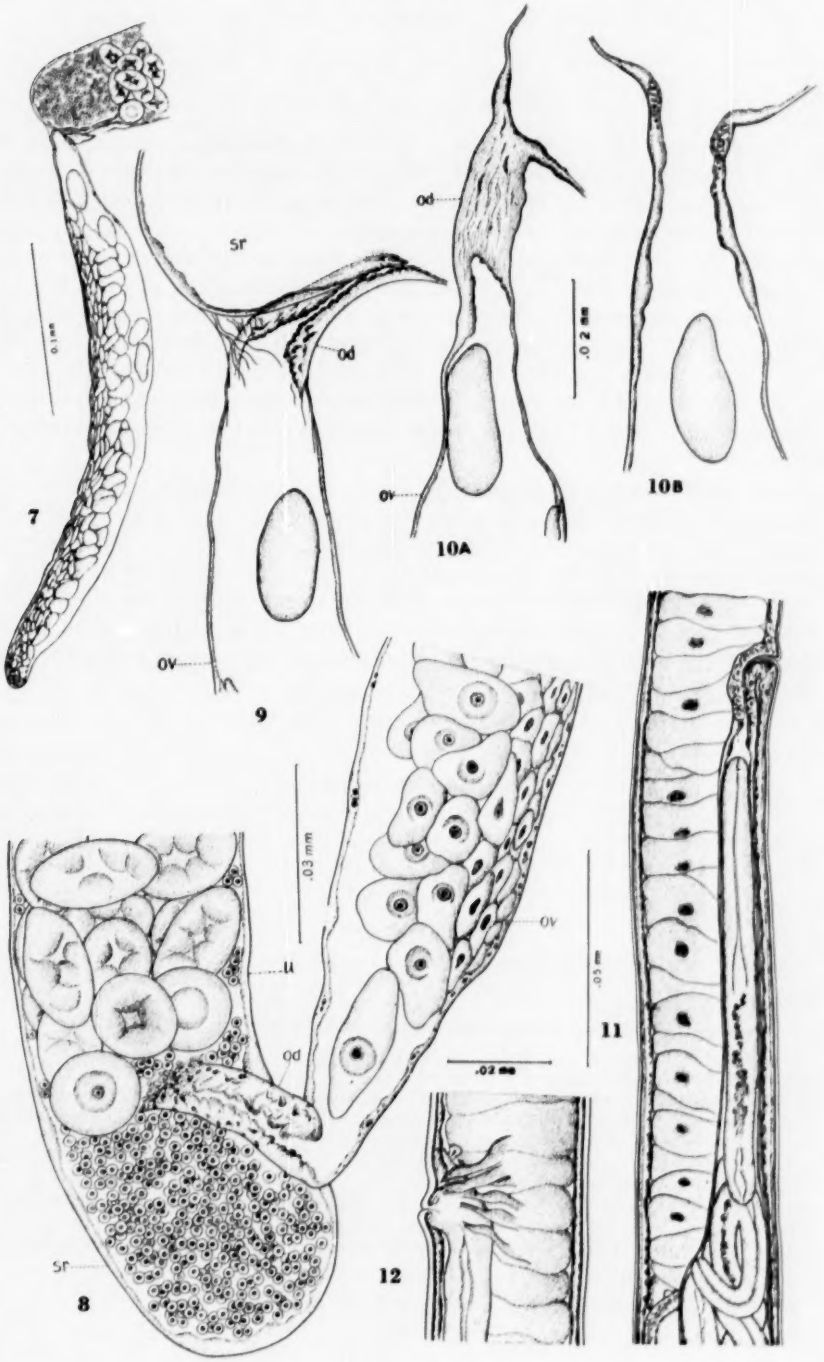


PLATE II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A NEW SPECIES OF *BENEDENIA* (TREMATODA: MONOGENEA) FROM *GIRELLA NIGRICANS*, THE OPALEYE

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Florida State University, Tallahassee*

Early in 1953 Dr. R. J. Menzies of the Scripps Institute of Oceanography at La Jolla, California forwarded a vial of monogenetic flukes to the writer for identification. This material had been collected earlier by Dr. Cadet Hand, also of Scripps Inst. (The writer wishes to thank both of these scientists for their aid.) These worms were studied and found to be a hitherto undescribed species of the genus *Benedenia* Diesing, 1858. The present paper is a description of this new species.

Judging from the color and condition of the preserved material it seems probable that the worms were fixed under slight pressure in Bouin's fluid. The following description is based on a large series of whole mounts stained with alum cochineal, hematein or Delafield's hematoxylin, cleared in deacidified beechwood creosote and mounted in piccolyte. Unstained whole worms mounted in Diaphane were also employed.

All measurements were made by the use of a calibrated ocular micrometer and are given in millimeters. In citing the measurements the mean is given, followed by the minimum and maximum in parentheses. The number of measurements used in deriving the mean is usually the same as the number of specimens measured, but where different the number employed appears in parentheses before the measurements. All measurements of curved structures are across the lines subtending the greatest arcs described by those structures. Drawings were made with the aid of the camera lucida.

Superfamily CAPSALOIDEA Price, 1936
Family CAPSALIDAE Baird, 1853
Subfamily BENEDENIINAE Johnston, 1931
Genus *Benedenia* Diesing, 1858

Benedenia girellae n. sp.

(Figs. A–J)

Host: Girella nigricans (Ayres), the Opaleye, a nerito-pelagic marine fish of the family GIRELLIDAE.

Location: Skin.
Locality: La Jolla, California.
Number studied: 39.
Number measured: 7.
Holotype (49,328) and paratype (49,329) in the USNM Helm. Coll.

Description: Body elliptical, 5.7 (3.8–6.7) long by 2.7 (1.9–3.2) wide. Cuticle fairly thick and smooth. Prohaptor a pair of ventro-lateral anterior suckers, 0.442 (0.357–0.510) in diameter. Opisthaptor a concavo-convex, oval sucker, about 1.1 (0.9–1.3) in diameter, opening ventrally; armed by 3 pairs of anchors, 14 marginal hooks and surrounded by a delicate marginal membrane. Anchors of first pair stout, conical to hastate, 0.271 (0.191–0.357) long, anteriorly directed; anchors of second pair slender, compressed, with recurved tips, 0.365 (0.261–0.427) long; anchors of third pair very slight, with depressed shafts and delicate recurved tips, 0.126 (0.070–0.166) long. Hooks delicate, sickle-shaped, 0.009 (0.007–0.011) long, in rim of muscular part of opisthaptor. Mouth midventral between anterior suckers; buccal canal short. Pharynx lobulated, about 0.430 (0.223–0.516) in diameter; esophagus not

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present. Gut bifurcated, crura ramified medially and laterally, rami dendritic, crura not confluent posteriorly. Two testes side by side, equatorial, lobulated, fenestrated, oval in outline, 1.054 (0.758–1.236) long by 0.602 (0.478–0.701) wide; vasa efferentia anastomosing to form sinuous vas deferens which parallels left intestinal crus on its course to cirrus. Cirrus complex consisting of muscular cirrus, prostate reservoir, and seminal vesicle in cuticularized cirrus pouch. Entire complex obliquely situated, posterior to pharynx, 0.702 (0.510–0.847) long by 0.176 (0.140–0.191) wide; seminal vesicle flask-like, prostate reservoir flask-like, both appearing to fuse distally to form a narrow intromittent organ. Prostate gland cells near excretory vesicles, ducts opening into opposite ends of the cirrus pouch. Glands of Goto postero-mesial to testes. Common genital pore submarginal on left near anterior level of pharynx, opening into the genital atrium. Ovary pretesticular, oval, containing seminal receptacle (as described for *Benedenia melleni* (MacCallum, 1927) Johnston, 1929) whose lumen often contains spermatozoa and ova; oviduct receives duct from vitelline reservoir at a small, dextrally placed, flask-shaped structure then proceeding anteriorly to ootype. Ootype wide, thick-walled, surrounded by many Mehlis' gland cells; uterus short, opening into genital atrium. Vitellaria follicular, situated in all parts of body near intestinal crura; transverse vitelloducts fusing medially to form an oblong vitelline reservoir immediately anterior to ovary; vitelline reservoir duct extends from anterior edge of reservoir to join oviduct posteriorly. Eggs polyhedral, (3) 0.270 (0.159–0.325) long by 0.219 (0.127–0.268) wide, long filament at one angle and short one at another. "Brain" dorsal to buccal canal, two pairs of anterior nerves and at least one pair of large, laterally located posterior nerves present. Four granular eyespots located near brain. Excretory ducts slightly lateral to and paralleling the intestinal crura; excretory vesicles antero-lateral to vitelline reservoir, opening via dorsal excretory pores.

Discussion: *Benedenia girellae* n. sp. is apparently most closely related to either *B. melleni* (MacCallum, 1927) Johnston, 1929 or *B. isabellae* Meserve, 1938; however, it differs from both in the following respects: (1) greater body size, (2) anchor sizes and shapes, (3) unusual junction of the vitelline reservoir duct and oviduct unique, (4) host family.

This description increases the number of avaginate species in the genus *Benedenia* to five. These species are *B. adenea* Meserve, 1938, *B. girellae* n. sp., *B. isabellae* Meserve, 1938, *B. melleni* (MacCallum, 1927) Johnston, 1929 and *B. muelleri* (Meserve, 1938) Price, 1939. Price (1939) suggested that a separate genus may have to be erected for the worms which feature this character. Although the absence of a vagina and slight differences in anchor shapes are probably only subgeneric in stature, the writer agrees that some taxonomic separation of these species from the other species of the genus is justifiable. If a supraspecific category is named for them, e.g. a subgenus, then Johnston's (1929) subgenera must be considered and integrated.

Literature Cited

Johnston, T. H. 1929 Remarks on the synonymy of certain tristomatid trematode genera. Tr. and Proc. Roy. Soc. South Australia **53**: 71–78.

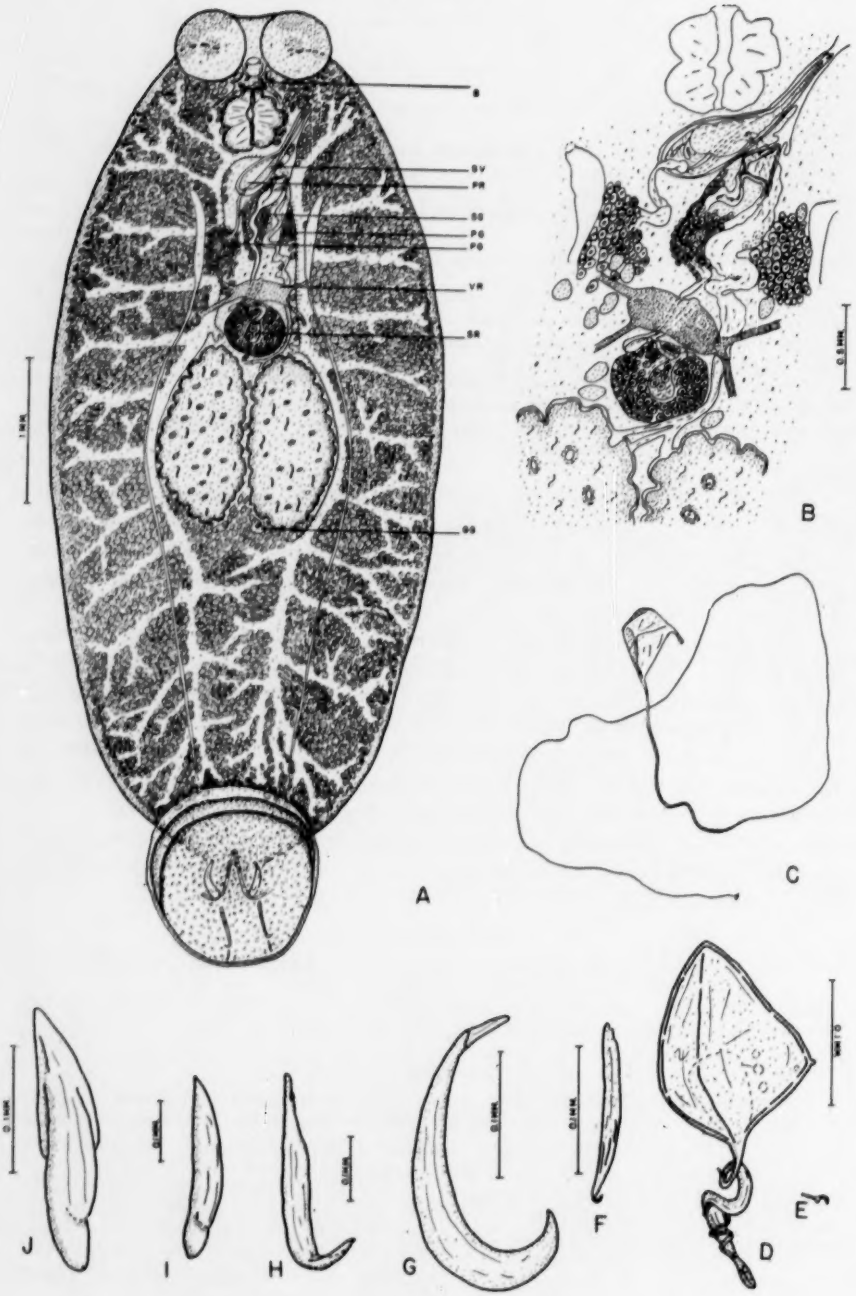
Price, E. W. 1939 North American monogenetic trematodes. III. The family Capsalidae (Capsaloidea). J. Wash. Acad. Sc. **29**: 63–92.

Explanation of Plate

Abbreviations: B "Brain," GG Gland of Goto, PG Prostate glands, PR Prostate reservoir, SG Mehlis' gland cells, SR Seminal receptacle, SV Seminal vesicle, VR Vitelline reservoir.

Fig. A. Whole mount of *Benedenia girellae* n. sp., ventral view.
Fig. B. Enlargement of male and female organs.
Fig. C. Freehand sketch of free egg.
Fig. D. Drawing of egg *in utero*.
Fig. E. Marginal hook.
Fig. F. Anchor of third pair.
Figs. G. and H. Anchors of second pair. Side and three-quarter views respectively.
Figs. I. and J. Anchors of first pair, showing different shapes.

PLATE I



THE IDENTIFICATION OF *TAENIA TENUICOLLIS* RUDOLPHI, 1819, IN NORTH AMERICA[1]

BETTY LOCKER

Taenia tenuicollis and its larval stage, originally named *Cysticercus talpae*, were described from European hosts by Rudolphi (1819). This tapeworm was first recognized as part of the helminth fauna of North America when Skinker (1935) reported the larvae, both cysticerci and coenuri, in the livers of muskrats in Ontario, Canada, and the adult cestode in mink from the same locale. She presented a review of the European literature and thus clarified the early nomenclature of this cestode. She also listed the first North American host records in muskrat and mink in Ontario, Canada. Since Skinker's paper, published records of this tapeworm's occurrence in the United States have been limited to a tentative diagnosis of the larvae in Michigan muskrats by Ameel (1942). Skinker's drawings of larval hook shapes corresponded closely with the adult and larval hooks found in this survey and were used as the basis for diagnosis of the larval cestodes.

Baylis (1935) reported the cysticercus from muskrat in Shropshire and Perthshire, England, where the North American muskrat had been introduced. Baylis, unaware of Skinker's record in Canada, postulated that the muskrats had acquired a European parasite after their introduction into England. He had also found the cysticercus in "*Evotomys glareolus, Microtus agrestis, Arvicola amphibius* and the grey squirrel (*Sciurus carolinensis*) in Great Britain, and in *Evotomys rufocanus* from Norway." Warwick (1936) also found the muskrat in Shropshire and Perthshire, England, infected with the larvae and records an instance of over 100 larvae in one liver.

Ameel (1942) found cestode larvae in muskrats from southern and central Michigan, which he recognized as similar to larvae of *T. tenuicollis*. However, he would not positively indentify them as such without feeding experiments to recover the adult cestode.

In Alaska, Rausch (1952) reported the intermediate stages from *Clethrionomys rutilus dawsoni, Lemmus trimucronatus alascensis, Microtus miurus pancaki;* and the adult cestode from arctic ermine, *Mustela erminea arctica*.

Joyeux and Baer (1936) listed a variety of European mustelid carnivores including ermine, weasel, pole cat, and marten as hosts of adult *T. tenuicollis*, and *Microtus arvalis* and *Cricetus cricetus* as intermediate hosts.

In addition to the above mentioned European hosts, Wardle and McLeod (1952) gave *Apodemus sylvaticus, Clethrionomys glareolus* and *C. rufocampus* [sic] as intermediate hosts; and *Mustela nivalis*, and *M. erminea* as final hosts. These authors listed Skinker's records from Canada and compared her data on the adult cestode with those from Europe.

This present paper records data which the author has accumulated in the last 3 years on various intermediate and final hosts of this tapeworm in Alaska, Montana, and Oregon.

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[1] From the U. S. Department of Health, Education, and Welfare, Public Health Service, National Institutes of Health, National Microbiological Institute, Rocky Mountain Laboratory, Hamilton, Montana.

METHODS

Cysticerci and coenuri were dissected from the liver tissues of the rodent intermediate hosts and fixed in aceto-formalin-alcohol. These cysts were either stained, dehydrated, and mounted in clarite, or the rostellum with hooks was dissected away from the scolex and mounted directly in polyvinyl alcohol. The adult worms from mustelids were relaxed in water, then fixed in aceto-formalin-alcohol, stained in Semichon's carmine, dehydrated, and mounted in clarite.

Identification of the larval stages was made by measurements of hook size and shape, and then comparison with Skinker's drawings and data.

NEW RECORDS OF THE LARVAL STAGES

Alaska: Livers of 7 muskrats collected near Beaver, Alaska, were frozen and air mailed to the Rocky Mountain Laboratory because of the observation of large white blotches on them. Inoculation of portions of these tissues into mice failed to demonstrate an infectious agent, and closer inspection revealed the helminth nature of the blotches. Of the 7 specimens received, 2 were grossly parasitized by coenuri of *T. tenuicollis*, 2 showed less severe infections, and the remaining 3 were not infected. Upon the diaphragmatic and visceral surfaces of one of the grossly parasitized livers, approximately 130 coenuri (each about 8 mm. in diameter) were visible (Fig. 1). Histological examination showed slight fibrotic liver tissue reaction and leucocyte infiltration surrounding the cestode capsule. A cross-section of this liver (Fig. 2) shows the gross morphology of the coenuri.

Two *Microtus oeconomus* were collected at a later date from Beaver, Alaska, and each had a liver cysticercus of *T. tenuicollis*.

Montana: Of 24 muskrats, *Ondatra zibethica osoyoosensis*, trapped in Ravalli

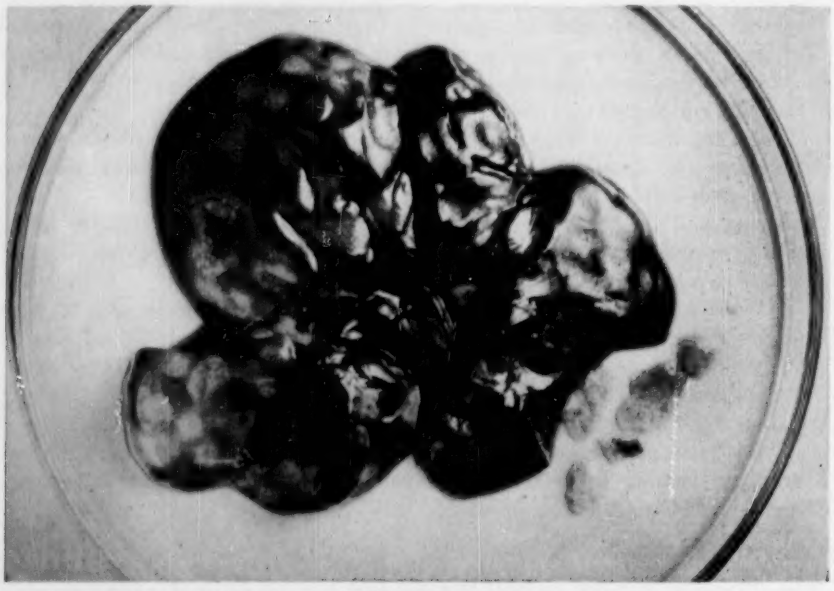

FIG. 1. Diaphragmatic surface of Alaskan muskrat liver showing cestode coenuri.

FIG. 2. Cross section of Alaskan muskrat liver showing random placement of cestode coenuri. ×3.

County during 1950, 3 had liver coenuri. During 1953, 97 muskrats from Ravalli County were examined, 14 of which were infected with *T. tenuicollis*. All of the parasitized animals came from one locale where 33 animals had been trapped. Each of these had from 1 to 3 coenuri in its liver. Of 99 muskrats from Lake County, none were parasitized with *T. tenuicollis*.

Of 90 *Microtus pennsylvanicus modestus* trapped in Ravalli County during 1950–52, 2 were parasitized, each with one cysticercus in its liver. These cysticerci were 2–2.5 mm. in diameter. Of 57 *Microtus pennsylvanicus* examined from Camas Creek, Glacier National Park, 9 had cysticerci and/or coenuri. The coenuri were 2–3 mm. in diameter.

Of 14 *Peromyscus maniculatus artemisiae* caught in Lincoln County in May 1953, one had a discrete pocket of 6 cysticerci in its liver. One *Microtus pennsylvanicus* from the same trapline contained a small coenurus with 11 scolices.

Oregon: During the winter and spring of 1951, 22 mountain beaver, *Aplodontia rufa*, were collected in the vicinity of Portland, Oregon, and 11 contained coenuri and cysticerci of *T. tenuicollis*. Some livers of these mountain beaver showed an amount of larval invasion comparable to the muskrat livers from Alaska, but instead of being only actively walled off from the liver tissue, these larvae in many instances were in various stages of calcification and fibrotic degeneration of a non-specific nature. One mountain beaver was immature so that its infection was of less than a year's duration, yet the cysts found in its liver also showed these same degenerative changes that appeared in older animals. This indicates that the larvae were consistently dying within the liver soon after establishing themselves there.

Thirty-three muskrats examined from the vicinity of Portland, Oregon, were without the larvae.

Of 35 *Peromyscus maniculatus rubidus* trapped in the Columbia Gorge near Multnomah Falls, Oregon, one had a cysticercus in its liver. Sections of this 1.5 mm. cyst revealed the typical hooks of *T. tenuicollis*. The cyst had been actively walled off by the host but showed no degenerative changes.

NEW RECORDS OF THE ADULT WORM

Montana: In Ravalli County, fragments of the adult cestode were recovered from an ermine, *Mustela erminea invicta*. In Flathead County, 55 adult worms (some with hooks) were recovered from 1 of 3 mink, *Mustela vison*. In Beaver-

head County, at the Red Rock Lake Refuge, adult worms were recovered from another mink, *Mustela vison*.

MORPHOLOGICAL AND TAXONOMIC CONSIDERATIONS

The general similarity of *T. tenuicollis* hooks to other taenioid hooks is obvious. However, careful measurements and comparison with hooks of *Cladotaenia* and other species, demonstrate differences sufficient to differentiate *T. tenuicollis*.

Hook characteristics differentiating the species of *Taenia* are among the most constant taxonomic criteria available at present, particularly when a large enough series of the species has been available for study to determine the limits of variability. In the author's experience it has been evident that certain of these small liver cysticerci from a wide range of hosts and localities had hooks which were consistent within a narrow size and shape range. Skinker (1935) has previously referred this group to the European species, *Taenia tenuicollis*.

During the accumulation of the preceding data, which are tabulated in Table I,

TABLE I.—*New Host Records for* T. tenuicollis *Rudolphi*, 1819

	Locality	Number examined	Number animals with			Number with adults
			cysticerci	coenuri	both	
Intermediate hosts						
Ondatra zibethica subsp.	Beaver, Alaska	7		4		—
O. zibethica osoyoosensis	Ravalli County, Montana	121		17		—
Microtus oeconomus	Beaver, Alaska	2	2			—
M. pennsylvanicus modestus	Ravalli County, Montana	90	3			—
M. pennsylvanicus modestus	Lincoln County, Montana	1		1		—
M. pennsylvanicus modestus	Glacier National Park	57	6	2	1	—
Peromyscus maniculatus artemisiae	Lincoln County, Montana	14	1			—
P. maniculatus rubidus	Multnomah County, Oregon	35	1			—
Aplodontia rufa	Multnomah County, Oregon	22			11	—
Final hosts						
Mustela ermina invicta	Ravalli County, Montana	3		—		1 (1 cestode)
M. vison	Flathead County, Montana	3		—		1 (55 cestodes)
M. vison	Beaverhead County, Montana	1		—		1 (several)

it has become apparent that the hook characteristics of this species are relatively constant, i.e., the hooks of both cysticerci and coenuri from various intermediate hosts are consistent with one another and with the hooks of the adult cestode from mink. This absence of any ambiguous shapes or sizes of hooks, while the larval body has expressed itself in two forms, substantiates Skinker's opinion that transitions between cysticercus and coenurus cysts among certain species of *Taenia* are commonly possible. She quoted European investigators who have found "proliferating" cysts of *T. tenuicollis* with "many scolices . . . in the [cestode] bladder." It may merely be an historical accident that the larval stage of *T. tenuicollis* was originally described as a cysticercus. Certainly among the muskrats, which were the most notably affected animals found during this work, the coenurus was the most common larval stage encountered.

The coenurus is from 4–10 mm. in diameter with as many as 120 scolices, and is easily seen at necropsy. The cysticercus in a small rodent's liver is 1–3 mm. in diameter and may be overlooked. When these small cysts are found they may,

if the hooks are not diligently looked for, be considered to be unarmed or designated as the larval stages of *Cladotaenia* spp. In all instances in which the larvae are fixed, stained, and cleared, the double row of typically taeniid hooks of distinctively small size (14–18 microns) and shape makes the species readily identifiable (*vide* Skinker, 1935). The adult worm in its mustelid host is one of the most delicately slender (60 mm. by 2 mm.) species of the genus, and occasionally retains a few of the characteristic hooks on its rostellum. Hook length in this survey ranged from 15.5–18.0 microns for the large hooks and 14.0–16.5 microns for the small hooks, which sizes are slightly smaller than similar ranges given by Skinker (16–20 microns and 16–18 microns, respectively). Also, scolices were never encountered with more than 48 hooks, 44 or 48 being the common numbers, whereas Skinker recorded as many as 60 hooks on a scolex.

SUMMARY

The list of intermediate hosts for *T. tenuicollis* is here extended to include other microtine rodents, *Microtus pennsylvanicus modestus* (in Montana), *M. oeconomus* (in Alaska), and races of the North American muskrat, *Ondatra zibethica* (in Alaska and Montana); two cricetine rodents, *Peromyscus maniculatus rubidus* (in Oregon) and *P. maniculatus artemisiae* (in Montana); and the mountain beaver, *Aplodontia rufa* of the relique rodent family Aplodontidae (in Oregon). On the basis of hook size, shape, and number, the larval types of *T. tenuicollis* are indistinguishable from one another even though, grossly, one form is a cysticercus and the other a coenurus.

The adult cestode is herein recorded from mink, *Mustela vison* (in Flathead and Beaverhead Counties, Montana); and from ermine, *M. erminea invicta* (in Ravalli County, Montana). The hooks of strobilae from the Flathead County mink are consistent with the hooks of larval forms; and the general morphology of these adult worms is the same as that of other strobilae from mustelids in Ravalli and Beaverhead Counties.

While the final confirmation of the specific identity or difference of these two larval types can come only from carefully controlled feeding experiments to compare adults reared from the two larval types, it is still suggestive that a single species is represented from the constant hook characters as manifested in the hooks of both the larval types from widely separate geographical areas, and the presence of both these larval types within the same species and twice within the same animal.

ACKNOWLEDGMENTS

The author wishes to acknowledge the help that many people have given both in field assistance and critical review. In particular, thanks should go to Major John M. Geary of the Biology Section, Aeromedical Laboratory, Ladd Field, Alaska, who air mailed to the Rocky Mountain Laboratory frozen muskrat livers which had been collected by Noel Ross near the village of Beaver, Alaska; to Clyde M. Senger and Kenneth A. Neiland who collected mountain beaver in the vicinity of Portland, Oregon, during the winter of 1951; Lowell Adams of the Fish and Wildlife Service who contributed material from his trapline in northern Montana; and to Dr. Philip Wright, University of Montana, who led the field work in Glacier National Park.

REFERENCES CITED

AMEEL, D. J. 1942 Two larval cestodes from the muskrat. Tr. Am. Micro. Soc. **61**: 267–271.

BAYLIS, H. A. 1935 Some parasitic worms from muskrats in Great Britain. Ann. and Mag. of Nat. Hist. **15**: 543–549.

JOYEUX, C. E. and BAER, J. G. 1936 Faune de France Cestodes, Féd. Franc. Soc. Sc. Nat. **30**, 613 pp.

RAUSCH, R. 1952 Studies on the helminth fauna of Alaska XI. Helminth parasites of microtine rodents—taxonomic considerations. J. Parasit. **38**: 415–444.

RUDOLPHI, C. A. 1819 Entozoorum synopsis cui accedunt mantissa duplex et indices locupletissimi. X + 811 pp., Berolini.

SKINKER, M. S. 1935 A redescription of *Taenia tenuicollis* Rudolphi, 1819, and its larva, *Cysticercus talpae* Rudolphi, 1819. Parasitology **27**: 175–185.

WARDLE, R. A. AND MCLEOD, J. A. 1952 The zoology of tapeworms. 780 pp., The Univ. Minn. Press, Minneapolis.

WARWICK, T. 1936 Parasites of the muskrat (*Ondatra zibethica* L.) in the British Isles. Parasitology **28**: 395–402.

AN ABNORMAL PROGLOTTID OF *TAENIA SAGINATA*

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Abnormalities occur frequently in cestodes and may involve the entire strobila or be limited to portions of it. Those affecting the entire strobila are: (a) triradiation (Clapham, 1939; and many others), tetraradiation (Foster, 1915) or pentaradiation (Rosenberger, 1903), in which the strobila shows three to five branches in cross section, the scolex generally possesses six to ten suckers and the rostellum may be doubled: (b) pigmentation, which may extend into every proglottid (Clapham, 1939); and (c) bifurcation of the entire strobila just posterior to the scolex (Chandler, 1930).

Malformations which affect one or more proglottids are: (a) bifurcation of a proglottid, which may, if equal, produce two similar chains of segments, or, if unequal, may produce two chains, the smaller of which gives the appearance of lateral branching (Blanchard, 1895); (b) fenestration, which is due to the absence of the central parenchyma of a proglottid (Moniez, 1890); (c) variations in segmentation, such as incomplete, oblique and spiral segmentation or absence of segmentation (Clapham, 1939); (d) supernumerary or intercalary proglottids, which are smaller, incompletely formed proglotids lying at the junction of two normal proglottids and which may fuse to one of the other proglottids (Foster, 1915; Moniez, 1890; Blanchard, 1895); (e) fusion of proglottids (McCulloch, 1913); and (f) variations in reproductive organs, such as, an increase or decrease in number or modifications in the structure of the organs (Roudabush, 1941; Honigberg, 1944).

OBSERVATIONS

An enlisted man who complained that he had been passing worms was instructed to bring in stool specimens, particularly any containing worms. One stool was received shortly after being passed and it contained 21 separate, but actively moving, proglottids. Among these was one abnormal specimen, which will be described below. Prior to treatment several other stools were brought to the laboratory with proglottids, none, however, being atypical. After treatment with oleoresin of aspidium, a complete *Taenia saginata*, including scolex, was passed.

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The broken chains when laid end to end measured 18 feet. A careful examination of the entire strobila failed to reveal other abnormal structures.

As soon as the abnormal proglottid was discovered, arrangements were made to have it photographed. The only camera available at the time was a fixed focus type and the proglottid is not in as sharp detail as was desired. However, the appearance of the contracted and expanded proglottid is shown in Fig. 1 A, B.

The proglottid was then flattened in an extended condition, fixed, stained and mounted (Fig. 1 C). Due to the torsion in the specimen, it folded to some extent, as shown near the upper right. Two genital atria (g) are present and from each extended a distinct vas deferens and a vagina. Although no line of demarcation could be found to indicate a fusion of two proglottids, the uterus appeared distinct

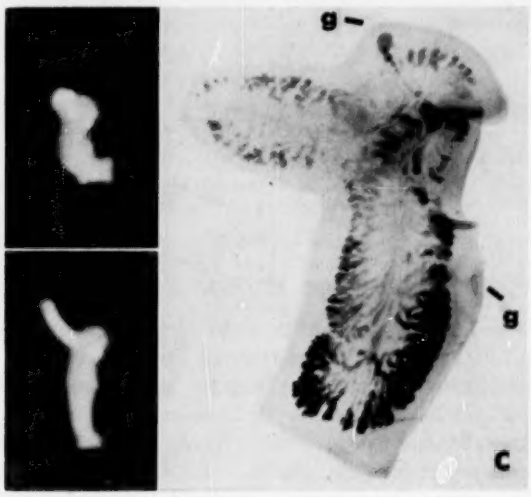

FIG. 1. An abnormal proglottid of *T. saginata*. A. Contracted. B. Extended. C. Flattened and stained. (A, B—Nat. size; C—×4).

in each portion. In the region where the uterine branches of the two systems meet (by upper fold) no fusion of branches could be found. With a binocular dissecting microscope the branches could be seen lying at different levels and overlapping each other, but not joining. The lateral excretory canals appear to be fused on the left, but cannot be made out distinctly on the upper right due to the fold.

When first seen, the proglottid had the appearance of an unequal dichotomy, but the stained specimen showed the presence of two complete reproductive systems. Therefore, it appears that the abnormality may be due to an early fusion of two unequal proglottids. The smaller one may have been a supernumerary proglottid that had fused to the normal one and although no external evidence of fusion could be found, the reproductive organs appear to have remained separate and distinct.

The authors have been unable to find reference to a previous case in which an exactly similar abnormality has been reported in Cestodes.

REFERENCES CITED

BLANCHARD, R. 1895 Sur un *Taenia saginata* bifurqué. Mém. Soc. Zool. Fr. **8**: 232–243.

CHANDLER, A. C. 1930 On a specimen of *Taenia pisiformis* with a completely double strobila. Tr. Am. Micr. Soc. **49**: 168–173.

CLAPHAM, PHYLLIS A. 1939 Some polyradiate specimens of *Taenia pisiformis* and *Dipylidium caninum*, with a bibliography of the abnormalities among cestodes. J. Helminth. **17**: 163–176.

FOSTER, W. D. 1915 Two new cases of polyradiate cestodes, with a summary of the cases already known. J. Parasit. **2**: 7–19.

HONIGBERG, B. 1944 A morphological abnormality in the cestode, *Dipylidium caninum*. Tr. Am. Micr. Soc. **63**: 342–344.

MCCULLOCH, H. 1913 Notes on cestode monstrosities. With a report of a new case of *Taenia saginata* with Y-shaped proglottides. Am. J. Trop. Dis. **1**: 453–461.

MONIEZ, R. 1890 Sul la bifurcation accidentelle que peut présenter la chaine des Cestodes et sur les anneaux dits surnuméraires. Rev. Biol. Nord France, Lille (1890–91), **3**: 135–142.

ROSENBERGER, R. C. 1903 Notes from the Pathological Laboratory of the Jefferson Medical College. A peculiar teratologic form of *Taenia saginata*. Am. Med. **6**: 93–97.

ROUDABUSH, R. L. 1941 Abnormalities in *Taenia pisiformis*. Tr. Am. Micr. Soc. **60**: 371–374.

THE ABUNDANCE AND DISTRIBUTION OF THE ECTOPARASITES OF THE HOUSE MOUSE IN MISSISSIPPI[1]

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Records of arthropods ectoparasitic upon the house mouse are not numerous in the literature. Fleas and mites have received the most attention, 9 species of fleas and 5 of mites having been recorded from the house mouse in the United States. The flea species are *Xenopsylla cheopis, Leptopsylla segnis, Nosopsyllus fasciatus, Echidnophaga gallinacea, Ctenocephalides canis, Orchopeas wickhami, Orchopeas leucopus, Doratopsylla blarinae,* and *Ctenophthalmus pseudagyrtes* (Stiles and Hassall, 1910; Ewing, 1929; Fox, 1940; Snell, 1941; Hubbard, 1947; Worth, 1950). Mites which have received mention in this connection are *Myocoptes musculinus* (LISTROPHORIDAE), *Myobia muris-musculi,* and *Radfordia affinis* (MYOBIIDAE), the tropical rat mite *Bdellonyssus bacoti,* and the rickettsialpox mite *Allodermanyssus sanguineus* (Stiles and Hassall, 1910; Ewing, 1929; Snell, 1941; Grant, 1942; Horn, 1949; Worth and Rickard, 1951). *Polyplax spinulosa* and *P. serrata,* the rat louse and the mouse louse, respectively, and *Hoplopleura acanthopus* are lice which have been taken from the house mouse (Stiles and Hassall, 1910; Ewing, 1929; Snell, 1941; Horn, 1949; Menzies, Eads and Hightower, 1951).

In connection with the murine typhus control program conducted in Mississippi by the State Board of Health and the Public Health Service Communicable Disease Center during the period July 1, 1949, to June 30, 1950, a survey of the house mouse and its ectoparasites was made in 45 localities located in 23 counties fairly well distributed geographically in the State. The counties and the localities surveyed are shown on the accompanying map.

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[1] This paper is adapted from a thesis presented to the Tulane University of Louisiana in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

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MATERIALS AND METHODS

House mice were trapped alive in cage traps of the type used in a rickettsialpox survey in New York City by Horn (1949). Trapping was conducted in business establishments, farm buildings, and residences. No mice were obtained from strictly feral situations. The traps were set in the late afternoon on 1 or 2 days of each week during the survey period. They were collected early the following morning and mice were transported to the laboratory in the traps. There they were killed and the ectoparasites removed by a brushing and washing technique as follows: the mouse was brushed vigorously from tail to head with a stiff brush. It was then soaked for 15 minutes in an aqueous solution containing 1 to 2 percent "Aerosol OT" wetting agent which received frequent shakings during the soaking period. The wetting solution and 2 rinses of fresh water were filtered to recover the ectoparasites. The filter paper was preserved in 70 percent alcohol. Later the parasites were washed from the paper and mounted in PVA medium for identification.

Early indentifications were made by Dr. Harry Pratt, Communicable Disease Center Laboratory, Chamblee, Georgia, and later ones by the author.

TABLE 1.—*Ectoparasites found on the house mouse in Mississippi July 1, 1949—June 30, 1950*

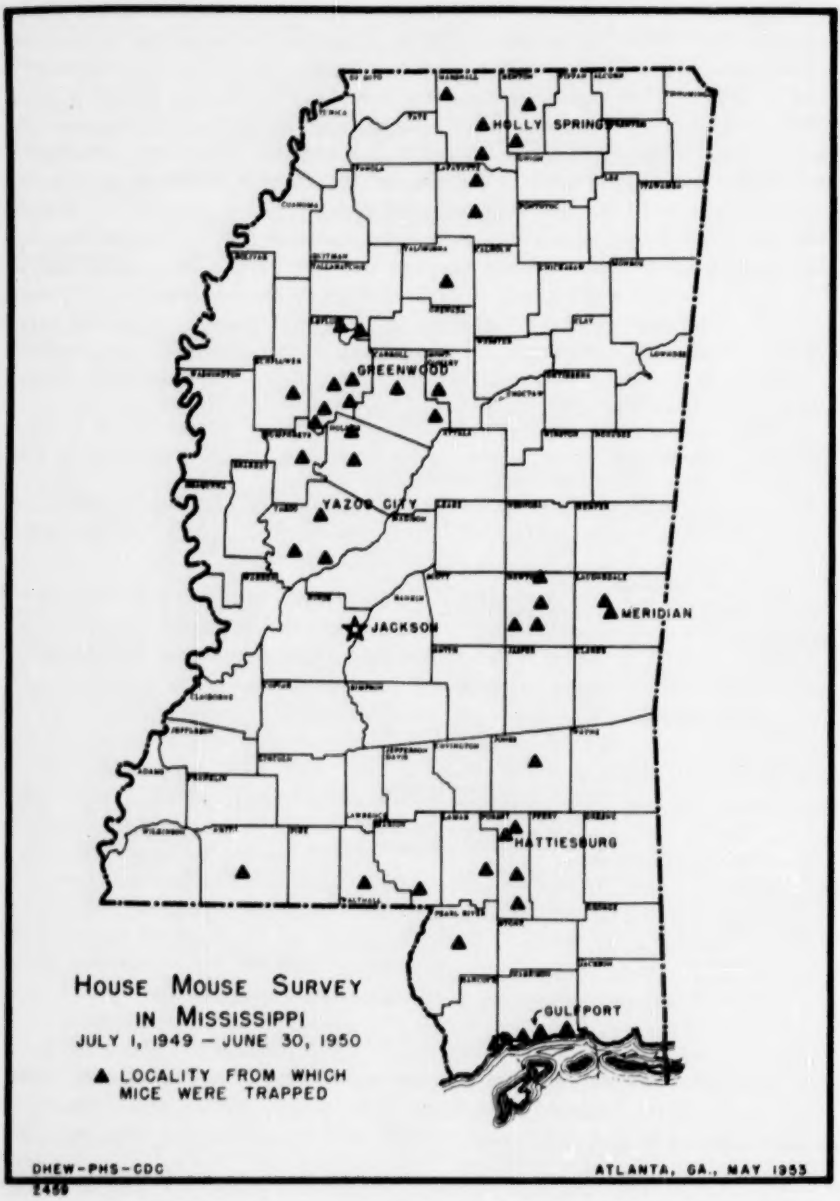
Family	Species	Ectoparasites		Mice	
		Total	Av. no. per inf. mouse	No. inf.	% Inf.
(Fleas—Siphonaptera)					
Pulicidae	*X. cheopis*	27	1.8	15	0.8
Dolichopsyllidae	*N. fasciatus*	13	1.9	7	0.4
Hystrichopsyllidae	*L. segnis*	31	1.4	22	1.3
Hectopsyllidae	*E. gallinacea*	1	1.0	1	0.1
		72	1.6	45	2.7
(Lice—Anoplura)					
Haematopinidae	*P. spinulosa*	29	1.0	28	1.7
(Mites—Acarina)					
Myobiidae	*Myobia muris-musculi*	3,297	4.4	752	45.4
"	*Radfordia affinis*	2,115	3.0	694	41.9
Listrophoridae	*Myocoptes musculinus*	3,621	5.9	615	37.1
Dermanyssidae	*Bdellonyssus bacoti*	760	3.0	256	15.5
"	*Ichoronyssus carnifex*	15	3.0	5	0.3
Laelaptidae	*Echinolaelaps echidninus*	10	1.7	6	0.4
"	*Laelaps nuttalli*	2	1.0	2	0.1
"	*Haemolaelaps glasgowi*	7	1.0	7	0.4
"	*Haemolaelaps mega ventralis*	1	1.0	1	0.1
Acaridae (Tyroglyphidae) spp.		277	1.5	187	11.3
Cheyletidae	*Cheyletus* sp.	11	1.0	11	0.7
Oribatellidae	*Oribatella* sp.	2	1.0	2	0.1
Uropodidae	*Uropoda* sp.	1	1.0	1	0.1
Phytoseiidae	Phytoseiinae sp.	1	1.0	1	0.1
Hypochthonidae	Hypochthoninae sp.	1	1.0	1	0.1
		10,134*	7.6	1,331	80.4
Survey Totals		10,235	7.6	1,344	81.2

* Includes 13 unidentified mites of the families Myobiidae and Listrophoridae.

ABUNDANCE DATA AND DISCUSSION

Four species of fleas, 1 species of louse, and at least 15 species of mites were found on the house mouse in Mississippi. The families, species, and numbers of ectoparasites taken from 1,344 mice are indicated in Table 1. A total of 1,656 mice was trapped, 312 being unifested.

It is to be noted that the fleas and lice were relatively scarce on the house mouse in Mississippi. The species listed are those which have been found most abundant on domestic rats in the southeastern United States by the PHS Communicable Disease Center Entomologic Services (1950). It is probable that these were ob-


HOLLY SPRINGS
GREENWOOD
YAZOO CITY
JACKSON
MERIDIAN
HATTIESBURG
GULFPORT
HOUSE MOUSE SURVEY
IN MISSISSIPPI
JULY 1, 1949 – JUNE 30, 1950
▲ LOCALITY FROM WHICH MICE WERE TRAPPED
DHEW-PHS-CDC
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ATLANTA, GA., MAY 1953

tained by the mice where their home ranges were overlapped by those of domestic rats. Other factors which may account for the small numbers of these arthropods are the tendency of the mice to rid themselves of the larger ectoparasites as observed by Worth (1950), and the reduction in numbers possibly caused by DDT dusting campaigns. The relatively small fur mites and the smaller listrophorid, *M. musculinus*, were most abundant. They tend to restrict themselves largely to areas of the body where it is difficult for the host to disturb or destroy them. In addition, the host is much more tolerant of these parasites, possibly due to their extremely small size (100–400 microns). The other mites are those commonly or occasionally found on domestic rats in Mississippi with the exception of *Ichoronyssus carnifex* (Koch). It has been reported by Hora (1934) to be associated with rodents in Lapland, Holland, Germany, and the Isle of Lewis (Great Britain). Its appearance is very similar to that of *B. bacoti*.

DISTRIBUTION

The following records indicate the counties in which each of the ectoparasite species was found on the house mouse.

Fleas

Echidnophaga gallinacea: Harrison
Leptopsylla segnis: Carroll, Forrest, Jones, Lamar, Lauderdale, Leflore, Marion, Newton Yazoo
Nosopsyllus fasciatus: Benton, Forrest, Marshall
Xenopsylla cheopis: Forrest, Holmes, Lafayette, Lamar, Lauderdale, Leflore, Marshall, Pearl River, Walthall, Yazoo

Lice

Polyplax spinulosa: Benton, Jones, Lafayette, Lamar, Marshall, Newton, Yalobusha, Yazoo

Mites

ACARIDAE (TYROGLYPHIDAE) spp.: Benton, Carroll, Forrest, Hancock, Harrison, Humphreys, Jones, Lafayette, Lamar, Lauderdale, Leflore, Marion, Marshall, Montgomery, Newton, Pearl River, Walthall, Yazoo
Bdellonyssus bacoti: Benton, Carroll, Forrest, Hancock, Harrison, Holmes, Humphreys, Jones, Lafayette, Lauderdale, Leflore, Marion, Marshall, Montgomery, Newton, Sunflower, Yalobusha, Yazoo
Cheyletus sp.: Carroll, Forrest, Harrison, Lafayette, Lauderdale, Leflore, Marshall, Yazoo
Echinolaelaps echidninus: Lauderdale, Marshall, Yalobusha, Yazoo
Haemolaelaps glasgowi: Benton, Leflore, Marshall, Yalobusha
Haemolaelaps megaventralis: Marshall
HYPOCHTHONINAE sp.: Leflore
Ichoronyssus carnifex: Leflore, Marshall, Montgomery
Laelaps nuttalli: Forrest, Newton
Myobia muris-musculi: Benton, Carroll, Forrest, Hancock, Harrison, Holmes, Humphreys, Jones, Lafayette, Lamar, Lauderdale, Leflore, Marion, Marshall, Montgomery, Newton, Pearl River, Sunflower, Yalobusha, Yazoo
Myocoptes musculinus: Same distribution as *M. muris-musculi* except not found in Walthall
Oribatella sp.: Lauderdale, Marshall
PHYTOSEIINAE sp.: Marshall
Radfordia affinis: Same distribution as *M. muris-musculi* except not found in Pearl River
Uropoda sp.: Forrest

SUMMARY

Four species of fleas, 1 species of louse, and at least 15 species of mites were found on house mice live-trapped from indoor habitats in Mississippi. These ectoparasites were obtained during a 1-year survey conducted in 45 localities of

23 counties well distributed throughout the State. The abundance and distribution of these ectoparasites are indicated. The mites were much more numerous than the other groups. Two fur mites, *M. muris-musculi* and *R. affinis*, and the listrophorid mite, *M. musculinus*, were most numerous and widely distributed. They are believed to be obligate ectoparasites of the house mouse. The tropical rat mite, *B. bacoti*, was the species of next importance in distribution and abundance. *I. carnifex*, a mite closely resembling *B. bacoti*, is recorded from the house mouse for the first time. The rickettsialpox mite, *A. sanguineus*, was not found present. The fleas, *X. cheopis*, *L. segnis*, *N. fasciatus*, and *E. gallinacea*, and the rat louse, *P. spinulosa*, which were also found on the house mouse in small numbers, are known to be ectoparasites of the domestic rat and are believed to have been obtained by the mice chiefly from this source.

ACKNOWLEDGMENT

Appreciation is gratefully expressed to Mr. H. A. Kroeze, State Director of Communicable Disease Center Activities, Division of Sanitary Engineering, Mississippi State Board of Health; Mr. H. M. Jones, Assistant State Director; Mr. H. M. Jamison, Asst. San. (R); and to the Communicable Disease Center field supervisors for their encouragement and assistance in this work.

REFERENCES CITED

EWING, H. E. 1929 A manual of external parasites. 225 pp. C. C. Thomas, Springfield.

FOX, I. 1940 Fleas of eastern United States. 191 pp. Iowa State College Press, Ames.

GRANT, C. D. 1942 Observations on *Myobia musculi* (Schrank) (Arachnida; Acarina: Cheyletidae). Microentomology **7**: 64–76.

HOBA, A. M. 1934 Notes on mites collected from the Isle of Lewis, Outer Hebrides. Parasitology **26**: 361–365.

HORN, M. B. 1949 Rickettsialpox survey in New York City. U. S. Public Health Service, Communicable Disease Center, Atlanta, Ga., CDC Bull. April, May, June, Atlanta, pp. 38–39.

HUBBARD, C. A. 1947 Fleas of western North America: their relation to the public health. 533 pp. Iowa State College Press, Ames.

MENZIES, G. C., EADS, R. B., AND HIGHTOWER, B. G. 1951 List of Anoplura from Texas. Proc. Ent. Soc. Wash. **53**: 150–152.

SNELL, G. D. 1941 Biology of the laboratory mouse. 497 pp. Blakiston Co., Philadelphia.

STILES, C. W. AND HASSALL, A. 1910 Compendium of animal parasites reported from rats and mice (Genus Mus.). [Pub. Health Bull.]. U. S. Health and Marine Hospital Serv. 1910, pp. 111–112.

U. S. PUBLIC HEALTH SERVICE, COMMUNICABLE DISEASE CENTER. 1950 ENTOMOLOGIC SERVICES. Murine typhus activities. CDC Bull. **9** (9): 16–17.

WORTH, C. B. 1950 House mice and commensal rats in relation to the dissemination of rat fleas. Amer. Jour. Trop. Med. **30**: 917–920.

——— AND RICKARD, E. B. 1951 Evaluation of the efficiency of common cotton rat ectoparasites in the transmission of murine typhus. Amer. Jour. Trop. Med. **31**: 295–298.

NEW HOST RECORDS FOR THE IMMATURE STAGES OF THE TICK *DERMACENTOR PARUMAPERTUS*

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Ecological Research, University of Utah

Published records indicate that animals of the family LEPORIDAE are the principal hosts for adult ticks of the species *Dermacentor parumapertus*. It is stated by Hooker, Bishopp and Wood (1912) that "while very few larvae and nymphs have been collected on rabbits, it would appear from the fact that no specimens have been taken on any other animal that the rabbit is the principal host of those stages." This belief is affirmed by Cooley (1938) who states "the only known hosts of the immature stages are rabbits."

In a study concerning the seasonal abundance of the tick, *D. parumapertus*, on the black-tailed jack rabbit (*Lepus californicus*), Fremling and Gastfriend (1954) found that the larval population on rabbits in the Great Salt Lake Desert in 1953–54 was insufficient to maintain the high nymphal and adult population found thereon. Hence, it follows that the larvae and possibly the nymphs must parasitize hosts other than rabbits.

In this region, *D. parumapertus* appears to occupy the lower valleys and deserts while *D. andersoni* is more or less restricted to higher elevations, although there may be some overlap between the two ranges. In general, specimens of *D. parumapertus* are rarely taken from man and in no case in our studies have they been found attached, whereas it is a common occurrence to find *D. andersoni* attached on man. Thus, it is assumed that all immature *Dermacentor* collected in the lower elevations of this locality belong to the species *D. parumapertus*. For a more positive identification approximately 200 larvae and 50 nymphs were reared in the laboratory using a kangaroo rat (*Dipodomys ordii*) for feeding by methods similar to those of Kohls (1937). All adults were found to be *D. parumapertus*.

To obtain an idea of the approximate distribution of ticks among the rodents, the records of collection of 590 specimens taken during the year 1952 were examined. It was found that 45 percent of all rodents taken during that year were infested with ticks. These ticks were not identified at the time, but later studies showed the presence of ticks of three genera, *Dermacentor, Ixodes* and *Haemaphysalis* thereon. A tabulation of these rodents by species is given in Table 1.

In a study of seasonal abundance of ticks (Fremling and Gastfriend, 1954), it was shown that 4,235 adults were of the species *D. parumapertus* and that 961 larvae and 2,984 nymphs of the genus *Dermacentor* must also belong to this species. Only 160 specimens belong to other species. These were taken from 197 rabbits during the period April 1, 1953 to March 31, 1954.

In order to study the occurrence of the immature tick population among the local rodents, the numbers of larvae and nymphs of the genus *Dermacentor*, undoubtedly of the species *parumapertus* found on 172 specimens of 12 species were counted. These were taken during the period August, 1951 to October, 1953 and are shown in Table 2.

* The author wishes to express his sincere appreciation to the staff of Ecological Research, University of Utah, for their assistance in this study.

TABLE 1.—*Rodent infestation with ticks for the period January 1 to December 31, 1952*

Hosts	No. examined	No. infested	% infested
Antelope ground squirrel (*Citellus leucurus*)	22	6	27.3
Chisel-toothed kangaroo rat (*Dipodomys microps*)	27	26	96.3
Ord kangaroo rat (*D. ordii*)	103	70	67.9
Kangaroo mouse (*Microdipodops megacephalus*)	26	15	57.7
Bushy-tailed wood rat (*Neotoma cinerea*)	2	2	100
Desert wood rat (*N. lepida*)	53	24	45.3
Long-tailed pocket mouse (*Perognathus formosus*)	32	7	21.9
Little pocket mouse (*P. longimembris*)	15	9	60
Great Basin pocket mouse (*P. parvus*)	29	3	10.3
Deer mouse (*Peromyscus maniculatus*)	244	89	36.5
Pinyon mouse (*P. truei*)	15	10	66.6
Western harvest mouse (*Reithrodontomys megalotis*)	22	5	22.7
TOTALS	590	266	45.0

DISCUSSION

From the evidence presented, it appears that immature stages of the tick, *D. parumapertus,* infest a wide variety of rodent hosts, a list of which is given. It is obvious from the tables that in this study the rodents were more heavily infested with larvae (average 4) than with nymphs (average 1.5) while in the rabbits, the trend is reversed, 961 larvae to 2,984 nymphs. Since adults are principally parasites of rabbits, it appears that in the studies made larval ticks are widespead in their choice of hosts, nymphs tend to concentrate more on rabbits than rodents, and adults concentrate on rabbits to the exclusion of rodents.

SUMMARY

In a study of 590 specimens representing 12 species of rodents, 266 (45 percent) were infested by ticks belonging to species of 3 genera. In a similar study of 172 specimens belonging to the same species of rodents, 671 larval and 260 nymphal ticks belonging to the species *Dermacentor,* probably *parumapertus,* were collected. In a study of the seasonal abundance of ticks on the black-tailed jack rabbit, *Lepus californicus,* it was shown that 4,235 adult ticks were of the species *D. parumapertus* and that 961 larvae and 2,984 nymphs of the genus *Dermacentor* must also belong to this species. Only 160 specimens belonged to other species.

As a result of these studies, it was found that animals of several species serve as hosts of the immature stages of *Dermacentor parumapertus.* Kangaroo rats belonging to two species of *Dipodomys* were the most heavily infested. Approximately three times more larvae than nymphs were collected from the rodent hosts,

TABLE 2.—*Infestations on new hosts of immature stages of* D. parumapertus

Host	No. of hosts infested	Specimens collected: Larvae		Nymphs	
		No.	Avg.	No.	Avg.
Citellus leucurus	4	2	0.5	7	1.7
Dipodomys microps	14	30	2.1	39	2.8
Dipodomys ordii	66	370	5.6	153	2.3
Microdipodops megacephalus	5	1	0.2	5	1.0
Neotoma cinerea	1	1	1.0	0	0.0
Neotoma lepida	4	14	3.5	11	2.9
Perognathus formosus	3	5	1.6	2	0.66
Perognathus longimembris	5	1	0.2	4	0.8
Perognathus parvus	3	3	1.0	1	0.3
Peromyscus maniculatus	57	229	4.0	36	0.6
Peromyscus truei	5	11	2.2	1	0.2
Reithrodontomys megalotis	5	4	0.8	1	0.2
TOTALS	172	671	4.0	260	1.5

whereas in the case of jack rabbits, three times more nymphs than larvae were found.

Literature Cited

Cooley, R. A. 1938. The genera Dermacentor and Otocentor (Ixodidae) in the United States, with studies in variation. Natl. Inst. Health Bull. No. 171, U. S. Publ. Health Serv. 89 pp.

Fremling, C. R., and Gastfriend, A. 1954. Seasonal abundance of the tick, *Dermacentor parumapertus*. (Unpublished report, Ecological Research, University of Utah.)

Hooker, W. A., Bishopp, F. C., and Wood, H. P. 1912. The life history and bionomics of some North American ticks. Bull. 106 U. S. Bur. Ent., U. S. Dept. Agric. 239 pp.

Kohls, G. M. 1937. Tick rearing methods with special reference to the Rocky Mountain wood tick, *Dermacentor andersoni* Stiles. In "Culture methods for invertebrate animals," Galtsoff, Lutz, Welch and Needham, Comstock Publishing Company Inc., Ithaca, pp. 246–256.

CHEMOTHERAPY OF AVIAN MALARIAS: III. TRIAL OF CERTAIN STANDARD ANTIMALARIALS ON *PLASMODIUM HEXAMERIUM* AND *P. ELONGATUM* MALARIA IN DUCKS*

Ikednachuku Okpala
and
Reginald D. Manwell

Since the species of malaria parasites differ widely in morphology, and somewhat in life history, it is logical that corresponding differences may exist, as has already been shown in various ways, among them, experiments in cultivation.

One may also expect to find differing responses to therapeutic drugs. Such peculiarities have been demonstrated for certain species, and for certain strains within species. But so far most species of malaria parasites have not been intensively studied from this point of view. We have, however, attempted it with *Plasmodium hexamerium* in ducks. This paper includes a report of additional work on *Plasmodium hexamerium*, and a beginning of similar studies of *P. elongatum*.

METHODS

Methods used were essentially those previously described (Manwell, Alpert and Powers, 1953; Manwell and Khabir, 1954), and the strain ("10D") of *Plasmodium hexamerium* was also the same. Our strain of *Plasmodium elongatum* however was a new one, without an official designation, originally isolated from a song sparrow (*Melospiza melodia melodia*) caught near Manlius, N. Y., on July 8, 1953. It has produced severe infections in ducks from the beginning.

The efficacy of drug therapy was measured primarily by 2 criteria: (1) ability of the drug to clear the blood of parasites, and (2) to produce sterilization (always confirmed by failures to relapse after splenectomy). In evaluating the suppressive power of each drug, or combination, asexual and sexual stages were tabulated separately in the counts.

The drugs used included camoquine, chloroquine plus quinine, and chloroquine plus primaquine, against *hexamerium* malaria, and camoquine, chloroquine and

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* From the Department of Zoology, Syracuse University.

primaquine for *elongatum* malaria. Details of treatment are given in Table 1. The dosage, which was larger on a body weight basis than that usually given man, was intentionally rather close to the maximum tolerated by the ducks, and was determined after considerable testing for toxicity.

Blood-induced infections were used, rather than mosquito-induced, because it is at present neither convenient nor practical to infect ducks otherwise with these species of plasmodia. The number of parasites in the inoculum was not rigidly controlled, although it was always large (100,000,000 to 200,000,000 in the case of *Plasmodium hexamerium;* 10,000,000 to 20,000,000 for *Plasmodium elongatum*). In any given experimental series, each duck received approximately the same parasite dosage, and this was roughly equivalent in the birds of all the series. The infective mixture was injected into the jugular vein.

Each experimental series consisted of 36 ducks: 1/3 were given their first treatment 12 hours after initial parasite inoculation, 1/3 beginning the 7th day, and 1/3 served as controls. In all, 216 ducks were used in experimental chemotherapy.

Since previous work had already shown *Plasmodium hexamerium* to be especially resistant to a number of standard antimalarials, a systematic search was also instituted for exoerythrocytic stages in the duck which might serve as a partial explanation. For this purpose, 58 ducks were infected and sacrificed in groups of from 1 to 3 daily, beginning 24 hours after initial infection and continuing for 25 days, by which time the parasitemia had usually reached a very low level.[1] Organ smears (brain, spleen, bone marrow, liver, lung, kidney and heart) were made, stained with J.S.B., and painstakingly searched.

RESULTS

Clearance of parasites from the blood was achieved in an average time of 3.6 days (Table 1), and 8 of the 24 treated birds were sterilized. Early treatment, as one might expect, was somewhat more effective than later therapy.

TABLE 1.—*Experimental chemotherapy of* hexamerium *and* elongatum malaria *in ducks*

Treatment		Stage of infection‡		Results		
Drug*	Dosage† (mg. per gm. body weight)	Early (number of birds)	Late (number of birds)	Parasite-clearance time (days: mean-range)	Sterilized number	Sterilized percent
A. *Plasmodium hexamerium*						
Camoquine	0.0294	12			5	41
			12	3.7 (1–5)	3	25
Chloroquine + quinine	0.06	12			3	25
	0.02		12	3.1 (1–4)§	1	8
Chloroquine + primaquine	0.06	12			4	33
	0.004		12	1.7 (1–3)	2	17
B. *Plasmodium elongatum*						
Camoquine	0.0294	12			2	17
			12	5.7 (5–7)	2	17
Chloroquine	0.06	12			2	17
			12	5.8 (4–7)	3	25
Primaquine	0.004	12			0	0
			12	3.8 (3–7)	1	8

* For the drugs we are indebted to the Sterling-Winthrop Research Institute and to the Research Department of Parke, Davis & Co. Drugs used were: camoquine hydrochloride, 1% solution; chloroquine diphosphate, 1% solution; quinine dihydrochloride, 1% solution; primaquine diphosphate, 0.2% solution.

† Dosage was oral, using a Luer syringe to which was attached a small calibre soft rubber tube (section of catheter). Ducks were weighed daily, and amount of drug given correspondingly adjusted. In all cases, two doses daily were given about 12 hours apart, for 7 days.

‡ "Early" = within 12 hrs. of infective parasite dose; "late" = 7th day after initial inoculation.

§ Treatment by this combination of drugs was relatively ineffective against the gametocytes. Figures given therefore refer only to the effect of therapy on asexual forms.

[1] The behavior of *Plasmodium hexamerium* in the duck has been described in detail by the junior author (Manwell, 1952).

Previous trials of chloroquine and of quinine did not completely eradicate the infection with this species of malaria. Both drugs were fairly good suppressants, however. When used together, the combination proved considerably better than either separately. Four of the 24 treated ducks were completely cured (sterilized), and early treatment was more effective than later. But the time required for clearing the blood of parasites was not significantly shortened, and gametocytes were little affected by therapy.

Primaquine, like quinine and chloroquine, when used alone against *hexamerium* malaria had proved relatively inaffective. Although it could abort the majority of infections when given early, it cleared the blood of parasites very slowly. How-

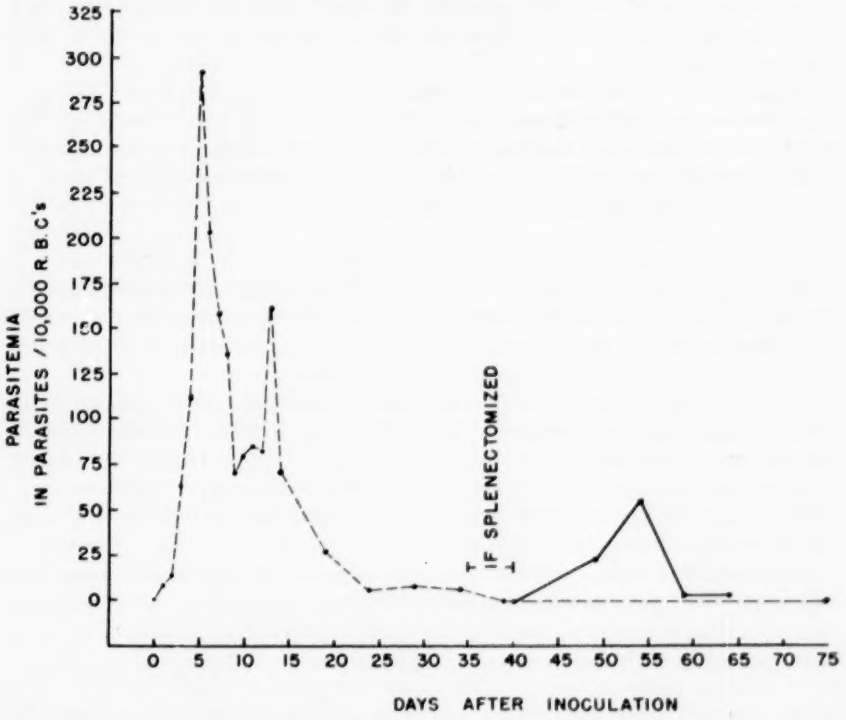

FIG. 1. Composite graph, based on counts made during the course of infection with *Plasmodium elongatum* in 36 ducks.

ever a combination of primaquine and chloroquine caused the rapid disappearance of the erythrocytic stages. But few cases (6 of 24) were sterilized, even though it required but 1.7 days of treatment to reduce the parasitemia to a point below the microscopically detectable level.

Search for exoerythrocytic stages as a possible explanation of the resistance of this species of avian plasmodium to chemotherapy again proved entirely fruitless. It is, of course, still entirely possible that they occur in the *Plasmodium hexamerium*-infected duck at some stage of the disease, but if so, their numbers must be small and it seems unlikely that they persist long.

Plasmodium elongatum, although it produces in ducks an infection with a lower

parasite level than *P. hexamerium*, is nevertheless more pathogenic. Due doubtless to a heavy concentration in the bone marrow, the resulting anemia is severe, and the disease often ends fatally. (In our experience with the present strain, the mortality was about 25 percent.) The course of parasitemia is shown in a composite graph (figure 1), based on parasite counts from 36 ducks.

Camoquine was somewhat less effective as a suppressant in *elongatum* malaria than when used against *Plasmodium hexamerium*, since it cleared the blood of parasites more slowly, but in only 4 of the 24 cases treated was there reason to believe that the infection was completely eradicated.[2]

Chloroquine also proved a weak drug against *elongatum* malaria. In almost half the cases treated (11 of 24) parasites were still easily demonstrable in stained blood films at the conclusion of treatment. Nevertheless, 5 of the remaining birds were sterilized.

Primaquine worked somewhat better. Moderately good parasite clearance from the peripheral blood was achieved, all but 3 of the 24 treated birds showing negative blood films at the conclusion of treatment. Yet in only 1 case was complete parasite eradication accomplished, thus making primaquine the weakest of the 3 drugs tested on this species of *Plasmodium*.

DISCUSSION

At this point, 2 questions may be asked. Is it worth while to test drugs against any species of animal malaria, when their potency in human malaria has already been demonstrated? Are results obtained on a *single strain* of any species of malaria (human or animal) likely to be significant?

To the authors, it seems highly desirable to obtain as much information as possible about both the therapeutic properties of given drugs, and the responses of given species to therapy. Such information gives a more detailed blueprint of what we know of various drugs and species, and it is necessary to fill in the gaps of the malaria picture, a picture which, from the biologists' point of view, is much larger than that of human malaria alone.

It is true that strains, in some cases with quite variant physiological characteristics, are known to exist within certain species, and perhaps such differing strains occur in most, or even in all species. But since it would be very difficult, and perhaps impossible, to isolate and test all such strains, experiments must usually be done on single strains, or on none. However there is no doubt that we know little of the extent to which such strains may exist, or how much they may differ, and there should be some careful investigation of this problem.

It seems at least reasonable to think that physiological differences among strains are likely to be less than between different species, which differ also in morphology and often in their life-histories.

If this is granted, it seems evident from the results presented here and in previous work, that drugs not only differ considerably in their ability to suppress or

[2] We believe that in these cases complete sterilization (in the sense that no parasites survived) was achieved. However Huff has pointed out (personal correspondence) that, in his experience with *Plasmodium elongatum*, persistence of the infection can occasionally be demonstrated by inoculation of bone marrow into healthy birds, even when all other signs are negative. Since we did not use this test, it is at least possible that some of our apparently "cured" cases of *elongatum* malaria may still have harbored parasites.

completely cure the various species of malaria, but that *Plasmodium hexamerium*, at least, is relatively resistant to nearly all the standard antimalarials. Of all the compounds, or drug combinations, so far tried against this species, only quinacrine (atebrine) proved both effective and relatively nontoxic. Pentaquine and isopentaquine (particularly the former) were quite effective in the dosage used, but both were also highly toxic. Neither camoquine, nor chloroquine used with either quinine or primaquine, equalled the effectiveness of pentaquine and quinacrine.

Continued inability to demonstrate exoerythrocytic forms in blood-induced infections of this species of malaria strongly suggests that they do not occur in the duck, and makes the high resistance of *Plasmodium hexamerium* to therapy even more remarkable. There is also the strong suggestion that, for this very reason, *hexamerium* malaria in ducks deserves serious consideration as a tool whenever it is desired to screen potential antimalarial compounds. Ordinarily trophozoite-induced malaria of any type is relatively more susceptible to chemotherapy, but sporozoite-induced infections are, in many laboratories, not practical, or less practical, for this kind of research.

The experiments so far done in the therapy of *Plasmodium elongatum* suggest that this species may be at least as resistant to treatment with the standard antimalarials as is *Plasmodium hexamerium*, and there is at least the likelihood that the 2 species will be found to differ considerably in their response to given chemotherapy. Camoquine sterilized a third of all the *hexamerium*-infected birds tested with it, but only one-sixth of those with *elongatum* malaria. Whether this should be regarded as a significant difference is difficult to say, for several reasons: The numbers of birds in the experimental series might well have been larger, the 2 species of malaria probably differ in their infectiveness (and the numbers of parasites in the inocula of the 2 species differed considerably), and there are peculiarities of life-cycle which separate them. *Plasmodium elongatum* tends to concentrate its attack on the bone marrow, whereas *P. hexamerium* seems confined to the peripheral blood. Strain differences within these species also no doubt exist.

It seems to the authors highly worthwhile that thorough testing of both known and future antimalarials be carried out on all species of plasmodia available for laboratory study. Why, for example, is camoquine, which has been found so effective in clearing the blood stream of parasites and dealing with the symptomatology of the various forms of human malaria, apparently much less effective against malaria in birds due to *Plasmodium hexamerium* and *P. elongatum*?

Only in ways such as this can adequate knowledge of the mechanisms involved in chemotherapy be finally obtained. After an accurate, and complete evaluation of each drug, the explanations of specific peculiarities of response can be sought in physiological differences between species of plasmodia, and between strains within species.

CONCLUSIONS

Studies of the effects of various standard antimalarials on blood-induced infections of *Plasmodium hexamerium* have been continued. Camoquine proved moderately effective in producing sterilization, and chloroquine plus primaquine rather quickly achieved parasite clearance from the peripheral blood. Chloroquine plus quinine, although it also cleared up the parasitemia fairly promptly, was the least

effective of the 3 regimens in achieving complete cure of *hexamerium* malaria. None equalled quinacrine or pentaquine.

Ducks infected with *elongatum* malaria (also blood-induced) were treated with camoquine, chloroquine, or primaquine. Each drug produced complete cure in 1 or more cases, but all were slow to clear the blood of parasites, and all failed to bring about sterilization of more than ¾ of the cases treated. Primaquine was the least effective of the three.

A very careful search for exoerythrocytic stages in *P. hexamerium*-infected ducks, made daily throughout the acute stage of the disease, failed completely.

REFERENCES

MANWELL, R. D. 1952 Turkeys and ducks as experimental hosts for *Plasmodium hexamerium* and *P. vaughani*. Exp. Parasit. **1**: 274–282.

———, ALPERT, S., AND POWERS, D. 1953 Experimental chemotherapy of *Plasmodium hexamerium* infections. J. Parasit. **39**: 209–214.

——— AND KHABIR, P. 1954 Further studies in the chemotherapy of *Plasmodium hexamerium* infections in ducks. J. Protozool. **1**: 105–110.

THE MODE OF EGG DISPERSAL IN *PHYSALOPTERA PHRYNOSOMA* ORTLEPP (NEMATODA: SPIRUROIDEA), A GASTRIC NEMATODE OF TEXAS HORNED TOADS, *PHRYNOSOMA CORNUTUM*

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Physaloptera phrynosoma is a common nematode in the stomach of Texas Horned Toads, *Phrynosoma cornutum*, and possibly of another horned lizard, viz. *Phrynosoma regale*. It occurred in more than three-fourths of the horned toads dissected in this study, many showing heavy infection.

P. phrynosoma was first described by Ortlepp (1922) on the basis of worms obtained from *P. cornutum* and *P. regale* that died in the London Zoological Garden. Ortlepp stated that these lizards were from Brazil but according to Ditmars (1946) and Günther (1902) the genus *Phrynosoma* inhabits only the arid part of the southwestern United States and similar climatic areas in Mexico. Harwood (1932) reported *P. phrynosoma* in *Phrynosoma cornutum* captured around Houston, Texas.

The genus *Physaloptera* is an ubiquitous group. Its primary hosts include amphibians, reptiles, birds, and mammals. The study of the life cycles of mammalian *Physaloptera* was initiated by Alicata (1937), then followed by Hobmaier (1941), Petri (1950), Petri and Ameel (1950), and Schell (1952). Only Petri and Schell completed the study of the entire cycle. The species of *Physaloptera* and their respective intermediate hosts studied by the above authors are listed below:

P. turgida—studied by Alicata (1937)
German cockroach (*Blattella germanica*)

P. maxillaris—studied by Hobmaier (1941)
German cockroach (*Blattella germanica*)

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[1] The author wishes to give his thanks to Dr. F. G. Wallace, Department of Zoology, University of Minnesota for advice in preparing this manuscript.

P. rara—studied by Petri and Ameel (1950)
German cockroach (*Blattella germanica*)
Flour beetle (*Tribolium confusum*)
Field crickets (*Gryllus assimilis*)
Ground beetle (*Harpalus* spp.)
P. praeputialis—studied by Petri and Ameel (1950)
German cockroach (*Blattella germanica*)
Camel crickets (*Centophilius* sp.) (emend *Ceuthophilus*)
Field crickets (*Gryllus assimilis*)
P. hispida—studied by Schell (1952)
German cockroach (*Blattella germanica*)
European earwig (*Forficula auricularia*)
Ground beetle (*Harpalus* spp.)

All the above authors obtained the mature embryonated and infected eggs from female worms taken from freshly sacrificed primary hosts. There is no extra membrane or capsule around the eggs of their respective worms. All the eggs are laid singly. All authors described three larval stages within the intermediate hosts.

The study of *Physaloptera phrynosoma* reveals the following features which are not found among the mammalian worms:

1. Unique mode of egg dispersal.
2. Egg capsules.
3. Development of larvae within the eggs.

MATERIAL AND METHODS

Horned toads were first kept in large wooden boxes with three to four inches of sand on the bottom and were fed daily with red ants (*Pogonomyrmex* sp.). Their droppings were collected daily and examined by sedimentation and floatation methods for eggs of *Physaloptera phrynosoma* but with negative results. Horned toads were sacrificed, therefore, in an attempt to obtain eggs but in two dozen animals dissected, only seven female worms with eggs were found though practically all the horned toads examined were infected with *P. phrynosoma* and many of them had over a thousand worms of various stages in their stomachs. Ortlepp also noticed the scarcity of mature females when he first described the species.

The eggs in these 7 females were not embryonated. They were given to German cockroaches, which were dissected 4 days later. Only egg shells were found in their hind guts.

One day the author by chance watched the process of defecation of one of the horned toads and saw one female *P. phrynosoma* being expelled separately from the fecal matter and still wriggling. The eggs of this worm were enclosed in separate capsules inside the uteri. Subsequent findings are the basis for the following description.

There are 5 to 69 eggs in each capsule, average 26. Many of the eggs in each egg capsule are empty. The fertile eggs are in various stages of development, from morulae to moving larvae.

At first the eggs are evenly distributed in both uteri then, gradually the eggs are gathered into separate masses through the peristaltic action of the uterine wall.

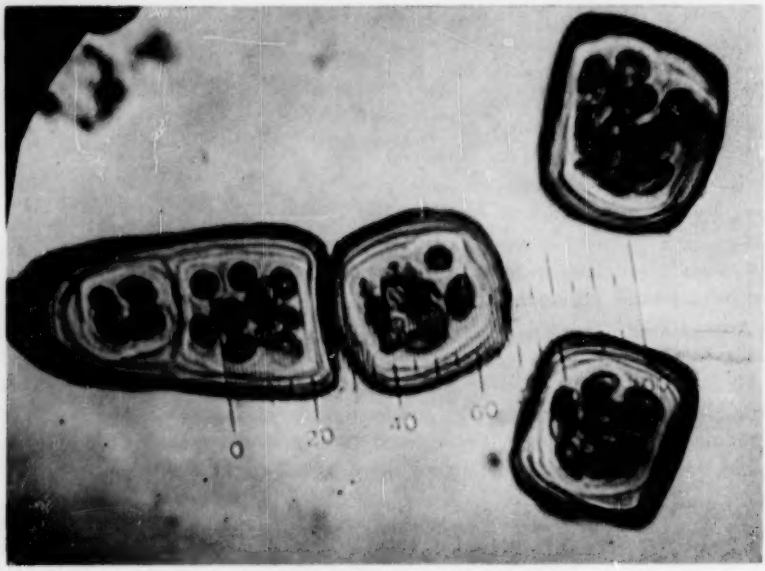

FIG. 1. Four isolated egg capsules. Each smallest division equals 0.0066 mm.

Five layers of sheaths are added to each mass through the secretion of the uterine epithelium. The sheaths of each capsule are of increasing thickness from the innermost layer outward. The thickness of the outermost layer is about 0.0018 mm.

The egg capsules are creamy to straw color when wet, but when dry they turn to pink orange. They range in mm. from 0.237 by 0.244 to 0.396 by 0.396. There is no definite shape, some are oval, some are round, some are cubic, and some are odd shaped with a length of 0.528 mm. (Fig. 1).

The eggs are oval, measuring from 0.061 by 0.053 mm. to 0.066 by 0.047 mm. The transparent covering around each egg measures from 0.011 to 0.0144 mm. thick. This covering disappears when the egg becomes dry, and does not reappear when the eggs are soaked in 0.8% saline or water.

Crushing the egg capsules under a cover glass releases many larvae through the broken egg shells and capsules. The larvae have an average length of 0.357 mm. and width of 0.022 mm. The esophagus appears as a short, 0.0104 mm., line. The excretory gland appears as a little triangle with its apex pointed toward the outside (Fig. 2).

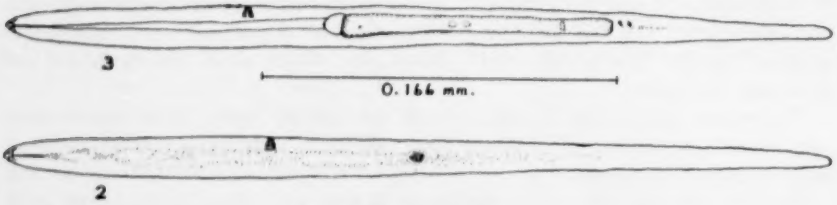

FIG. 2. First stage larva.
FIG. 3. Second stage larva.

The eggs from these freshly expelled worms were given to red ants, German cockroaches, Tribolium beetles and varied carpet beetles, *Anthrenus verbasci*. All these were opened within 2 weeks after feeding and all were found negative for nematode larvae.

Efforts were made to obtain females with egg capsules by dissecting the horned toads but they were found scarcer than females with eggs. It then appeared that the place to look for the females with egg capsules was not the stomach of the horned toads but in the boxes where the horned toads are kept. So the author began to keep his horned toads in empty cardboard boxes. Their droppings were dumped on a large sheet of black poster board weekly for easy discovery of worms. Two boxes housing ten horned toads each yielded two to three dry females every week. These dry females will not break unless deliberate effort is exerted, nor will they explode when broken. Under the stereoscopic microscope the uterine side has an orange color which disappears when the egg capsules inside become wet.

Embryonated eggs can remain viable over a year inside the uterus under dry conditions at room temperature, but deteriorate within a week if kept in water at room temperature and within 17 days in the refrigerator.

Worms collected on August 28, 1953, were examined on December 19, 1953 when second stage larvae were found in the egg capsules. These differ from the first stage larvae chiefly by having a well differentiated intestinal rudiment (Fig. 3).

Dry females were placed in a moist chamber at room temperature but no hatching or further development was observed.

DISCUSSION AND SUMMARY

This study indicates that the self-sacrificial exodus from the definitive host of *P. phrynosoma* is indispensable in its course of development. This mode of transmission has not previously been described. The female *Enterobius* may have the same destiny, but the essential nature of its sacrifice is not as acute as it is in *P. phrynosoma* for the *Enterobius* can discharge its eggs without emerging from the host and furthermore, when it does emerge it may expel its eggs while it is still alive or it may explode when dry. There is no dependence of eggs on the female *Enterobius* outside of the host. In *P. phrynosoma* the females continue to serve their eggs as protective coverings outside of the host. The mode of egg dispersal of *P. phrynosoma* evidently represents one extreme of parental care among helminths.

The egg capsule of *P. phrynosoma* is unique among all the nematodes. Similar structures are encountered among some cestodes, but their membranes are always thin. The egg capsule of *P. phrynosoma* on the contrary has 5 distinct membranes and apparently is relatively impermeable, thus helping to conserve water in the eggs.

The extra-arthropodal or free living developmental stages described in this study have never been found among other physalopterine worms.

ADDENDUM

Since reading the proof of this paper certain articles have come to my attention which indicate that the correct generic name for the nematode discussed above is *Skrjabinoptera* and that the correct name of the parasite is *Skrjabinoptera phrynosoma* (Ortlepp) Schulz, 1927.

REFERENCES CITED

ALICATA, J. E. 1937 Larval development of the spirurid nematode, *Physaloptera turgida*, in the cockroach, *Blatella germanica*. Rabot. Gelmint. (Skrjabin), pp. 11–14.

DITMARS, R. L. 1946 Reptiles of the World, Doubleday and Co., Inc., Garden City, N. Y.

GÜNTHER, A. C. L. G. 1902 Reptilia, Batrachia Biologia Centrali-Americana pp. I–XX, 301–325. London. Published for the editors of R. H. Porter, 10 Chandos St. Cavendish Square, W., and Dulau and Co., Soho Square W.

HARWOOD, P. D. 1932 The helminths parasitic in the Amphibia and Reptilia of Houston, Texas and vicinity. Proc. U. S. Nat. Mus. **81** Art. 17, pp. 1–71.

HOBMAIER, M. 1941 Extramammalian phase of *Physaloptera maxillaris* Molin, 1860 (Nematoda). J. Parasit. **27**: 233–235.

ORTLEPP, R. J. 1922 The nematode genus Physaloptera Rud. Proc. Zool. Soc. London, pp. 999–1107.

PETRI, L. H. 1950 Life cycle of *Physaloptera rara* Hall and Wigdor, 1918 (Nematoda: Spiruroidea) with the cockroach, *Blatella germanica*, serving as the intermediate host. Tr. Kansas Acad. Sc. **53**: 331–337.

——— and AMEEL, D. J. 1950 Studies on the life cycle of *Physaloptera rara* Hall and Wigdor, 1918 and *Physaloptera praeputialis* Linstow, 1889. J. Parasit. **36**: (6 Sect. 2): 40.

SCHELL, S. 1952 Studies on the life cycle of *Physaloptera hispida* Schell (Nematoda: Spiruroidea) A parasite of the cotton rat (*Sigmodon hispidus littoralis* Chapman). J. Parasit. **38**: 462–472.

NOTES ON FOUR HYMENOLEPIDID CESTODES FROM SHREWS

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Examination of a large number of shrews from California yielded tapeworms belonging to several known species of *Hymenolepis*. While identifying these cestodes, discrepancies between some of the California material and the original descriptions were noted. These observations, as well as a few new host and locality records are presented below. Further host and locality records are being presented elsewhere (Voge, In Press).

Appreciation is expressed to Dr. Robert Rausch, Arctic Health Research Center, Anchorage, Alaska, for the loan of shrew cestodes from Oregon and Alaska; and to Dr. Betty S. Davis, Hastings Reservation, Monterey County, California, for collecting some of the material discussed here.

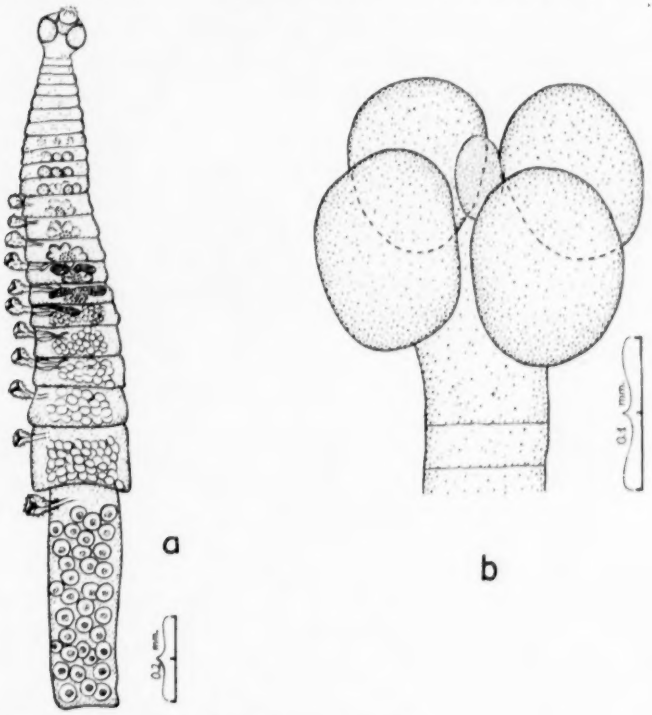
Hymenolepis macyi Locker and Rausch, 1952

Specimens obtained from two *Sorex trowbridgei*, 7 mi. E. of Bayside, Humboldt County, and 12 mi. S. of Korbel, Humboldt County, California, have a small, unarmed rostellum. Since *H. macyi* was originally described as having no rostellum, specimens from the type host and type locality were reexamined. In one of five specimens of *H. macyi* from *Sorex vagrans*, Portland, Oregon, (type host and locality) a small, unarmed rostellum was observed (Fig. b), measuring 30 by 54 micra; in the specimens from *Sorex trowbridgei*, this structure measured 22–32 by 40–50 micra. Failure to observe the rostellum in all specimens is due to the relatively small size of this structure which is usually obscured by the prominent suckers. In all other respects, my observations on this species are in accord with those of Locker and Rausch (1952).

Received for publication, July 2, 1954.

Hymenolepis sphenomorphus Locker and Rausch, 1952

Several specimens were obtained from *Sorex vagrans* collected at Red's Meadow, Madera County, California. As the original description of this cestode (Locker and Rausch, 1952) was based on specimens which lacked terminal gravid segments, a whole specimen from *Sorex vagrans,* with the very elongate terminal gravid segment is illustrated in Fig. a. Eggs in the terminal gravid segment are larger than reported by Locker and Rausch (1952); they measure 32–34 by 44–46 micra. The knob of the cirrus in terminal segments is larger than in more anterior proglottids and measures about 38 micra in diameter. In all other respects, the specimens from



Drawings were made with the aid of a camera lucida.
FIG. a. *Hymenolepis sphenomorphus.* Whole specimen with terminal gravid proglottid.
FIG. b. *Hymenolepis macyi.* Scolex with unarmed rostellum.

California conform to the original description of this species. Recently, Dr. Rausch (personal communication) has obtained *H. sphenomorphus* with terminal gravid segments from *Sorex* sp., collected at Anchorage, Alaska.

Hymenolepis kenki Locker and Rausch, 1952

This species was found in two *Sorex pacificus* collected 12 mi S. of Korbel, Humboldt County, California. Comparison of California material with the type specimen revealed no significant structural differences. The specimens from California are somewhat larger, measuring up to 3.4 mm. in total length and up to

420 micra in greatest width. So far as can be determined, the gravid segments in this species are considerably wider than long; their shape does not differ significantly from that of mature proglottids.

Hymenolepis falculata Rausch and Kuns, 1950

Originally described from *Sorex cinereus* by Rausch and Kuns (1950) in Wisconsin, this species was found in *Sorex palustris* and *Sorex vagrans*, Red's Meadow, Madera County, California, and in *Sorex ornatus*, Skyline Boulevard, San Mateo County, California.

SUMMARY

Four species of hymenolepidid cestodes are reported from shrews in California; *Hymenolepis sphenomorphus* from *Sorex vagrans; H. macyi* from *S. trowbridgei; H. kenki* from *S. pacificus;* and *H. falculata* from *S. palustris, S. vagrans* and *S. ornatus*. The host and locality records given above are all new. *H. sphenomorphus* is also reported from *Sorex* sp., Alaska.

An unarmed rostellum is described for *H. macyi* and the diagnosis of this species is thereby modified. The terminal gravid proglottids of *H. sphenomorphus* are described.

REFERENCES

LOCKER, B. AND RAUSCH, R. 1952 Some cestodes from Oregon shrews, with descriptions of four new species of Hymenolepis Weinland, 1858. J. Wash. Acad. Sci. **42**: 26–31.

RAUSCH, R. AND KUNS, M. L. 1950 Studies on some North American shrew cestodes. J. Parasit. **36**: 433–438.

VOGE, M. *In Press.* A list of cestode parasites from California mammals. Amer. Midl. Nat. —.

OBSERVATIONS ON THE BIOLOGY OF THE ARGASID TICK *ORNITHODOROS PUERTORICENSIS* FOX

GORDON E. DAVIS[1]

When Fox (1947) described *Ornithodoros puertoricensis* as a new species, he stated: "No data have been obtained as yet on copulation, fecundity, hatching time, larval engorgement time, and frequency of adult feeding."

On May 7, 1946, three engorged larvae were received from Dr. Fox and on June 10, 1946, three early stage nymphs were received. From these, a thriving colony has been established. The present report supplies data on the points mentioned above, and other information.

Throughout these studies, all ticks were stored at room temperature, in a closed jar, in which a relative humidity of 79 percent was maintained by a saturated solution of ammonium chloride.

Copulation:

Males seek the engorged females readily. The observed mating time for four pairs was 8, 8, 9, and 9½ minutes, respectively.

Received for publication, June 30, 1954.

[1] From the U. S. Department of Health, Education, and Welfare, Public Health Service, National Institutes of Health, National Microbiological Institute, Rocky Mountain Laboratory, Hamilton, Montana.

Intervals between Engorgement and Oviposition:

The intervals observed for eight ovipositions were: 7, 8, 9, 10, 11, 14, 16, and 26 days, respectively.

Oviposition:

Oviposition takes place rapidly. At one observation, there were two eggs in the early morning and by late afternoon the eggs were too numerous to count.

At times, there are two successive ovipositions without further feeding. This phenomenon has been noted especially in *O. nicollei* (Davis, 1943), in which there is quite regularly one oviposition in the spring and a second in the autumn without an additional blood meal or without further mating. In *O. puertoricensis*, the second oviposition takes place at about the time the first batch of eggs has hatched.

Intervals between Oviposition and Hatching:

The intervals observed at nine ovipositions were: 13, 14, 15, 15, 15, 15, 16, 21, and 22 days, respectively.

Fecundity and Fertility:

Egg and larva counts were made for eight ovipositions as follows:

Female no.	Eggs	Larvae	
46	182	174	First oviposition
46	108	96	Second oviposition, no further feeding or mating
38	249	168	First oviposition
38	426	332	Second oviposition, additional feeding and mating
1	148	114	First oviposition
3	140	133	First oviposition
3	352	343	Second oviposition, additional feeding and mating
5	191	138	First oviposition

Larval Engorgement Time and Recoveries:

In our early studies, the larvae were placed within a feeding capsule on the clipped belly of a guinea pig. Of 430 larvae thus placed in groups of from 10 to 53, using 17 guinea pigs, there was a recovery of 38.6 percent. However, in numerous attempts with uncounted lots of larvae, there were no recoveries. Of 371 engorged larvae recovered, from numerous guinea pigs, six required four days for engorgement; 132, five days; 163, six days; 51, seven days; and 19, eight days.

More recently, the guinea pig with the feeding capsule already attached, was strapped ventral side down on a board. The vial containing the larvae was then inverted over a clipped area on the back, within the capsule and held firmly until many of the larvae were attached. Using this method, the following results were obtained with two uncounted lots of larvae:

Guinea pig 1.		
Fourth day,	84	engorged larvae found detached at 9:00 am.
	44	engorged larvae found detached at 1:45 p.m.
Fifth day,	149	engorged larvae found detached at 9:00 am.
	29	engorged larvae found detached at 2:00 p.m.
Sixth day,	14	engorged larvae found detached at 9:00 am.
	33	engorged larvae found detached at 2:00 p.m.
Total	353	
Guinea pig 2.		
Fifth day,	115	engorged larvae detached
Sixth day,	23	engorged larvae detached
Seventh day,	25	engorged larvae detached
Total	163	

Of the 353 engorged larvae from guinea pig No. 1, all but 16 survived both larval and first nymphal molts. Fourteen of these were among the last recovered and most of them were obviously injured.

Rearing Data:

As a series of ticks had been reared from the larval stage to the adult stage before the publication by Fox, the data are presented here. There is complete agreement as to the number of nymphal stages but the molting time varied considerably between the two series, probably due to the different conditions in Puerto Rico and this Laboratory.

The time required for the larval molt was from 4 to 10 days. (1 in 4 days, 2 in 5 days, 28 in 6 days, 17 in 7 days, 3 in 8 days, and 1 in 10 days [52 ticks]). Without further feeding, the ticks molted to the second nymphal stage in from 16 to 31 days (45 ticks). The second stage nymphs molted to the third stage in from 17 to 20 days (26 ticks). The third stage nymphs molted to fourth stage nymphs or adults in from 20 to 48 days (22 ticks), and the fourth stage nymphs molted to adults, all females, in from 23 to 55 days (9 ticks). There were 12 males and 10 females. All males and one female appeared following the third nymphal molt, while the remaining nine females required an additional molt. Only a part of the original series was carried through to the adult stage.

Longevity:

Ticks fed as larvae in October 1946 are still alive in the adult stage as of May 1954.

Relation to Relapsing Fever Spirochetes:

Relapsing fever has not been reported from Puerto Rico and spirochetes have not been recovered from *O. puertoricensis.* Fox stated that there was no evidence that these ticks feed on man, however, they do feed readily (accidental) on man in the laboratory.

As a part of the tick-spirochete specificity studies carried on, the following two experiments were performed.

Experiment 1.

Ten *O. puertoricensis* and 10 *O. dugesi,* both in the second nymphal stage, engorged on a mouse infected with *O. dugesi* spirochetes. Over three test feedings, there was no transmission by *O. puertoricensis,* while there was transmission by nine of the *O. dugesi.*

Experiment 2.

Ten *O. puertoricensis* and 10 *O. talaje* (Kansas stock), both in the second nymphal stage, engorged on a mouse infected with Kansas *O. talaje* spirochetes. Over four test feedings, there was no transmission of *O. puertoricensis,* while there was transmission by nine of the *O. talaje.*

These results indicate a marked biologic difference within these three species of ticks.

Interbreeding Experiments:

Fox has suggested the possibility that the tick known as *O. talaje* in Panama is none other than *O. puertoricensis.* In this connection, it is of some interest that in this laboratory, *O. puertoricensis* and *"O. talaje"* from Colombia have interbred and produced fertile progeny. Further biologic studies on the three closely related "species," *O. puertoricensis, O. talaje,* and *O. dugesi* are in progress.

SUMMARY

Observations on the biology of *Ornithodoros puertoricensis* are reported in relation to copulation, intervals between engorgement and oviposition, oviposition, fecundity and fertility, intervals between oviposition and hatching, larval engorgement time and recoveries, rearing data, longevity, relation to relapsing fever spirochetes, and interbreeding.

REFERENCES

DAVIS, G. E. 1943 Studies on the biology of the argasid tick, *Ornithodoros nicollei* Mooser. J. Parasit. **29**: 393–395.

FOX, I. 1947. *Ornithodoros puertoricensis*, a new tick from rats in Puerto Rico. J. Parasit. **33**: 253–259.

STUDIES ON THE HELMINTH FAUNA OF ALASKA. XXIII. SOME CESTODE PARASITES OF EIDER DUCKS

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The 4 species of eider ducks, representing 3 genera, have circumboreal distribution. During the warmer months they occur as migrants or breeding birds in arctic Alaska and great numbers of the 2 more common species are killed by the Eskimo for food.

This survey was undertaken to determine the identity of cestodes obtained from eider ducks collected in Alaska during the past 5 years. The 4 species of ducks are: Steller's eider, *Polysticta stelleri* (Pallas); Pacific eider, *Somateria mollissima v-nigra* Gray; king eider, *Somateria spectabilis* (L.); and spectacled eider, *Arctonetta fischeri* (Brandt). In addition to the birds taken by the writer, this report includes those collected by Dr. Robert Rausch, of this laboratory. A list of the cestodes identified and data on prevalence, host numbers, and collection localities are given in Table 1.

The literature contains reports of a number of cestodes from some of the eider ducks; however, many of the early descriptions are so incomplete that recognition of the species is diffcult or uncertain. Relatively few of these species have been recorded subsequent to their original descriptions. Insofar as the writer has been able to determine, there are no previous records of cestodes from Steller's eider. The present study has yielded additional information on the morphological characters of some of these poorly known species. On the basis of the Alaskan material, 2 species are redescribed herein and 2 previously unknown species are described as new.

Haploparaxis polystictae n. sp.

(Figs. 1–3, 17)

(Measurements in microns, unless otherwise designated.)

Diagnosis: Length of strobila about 12 mm.; maximum width 450. Scolex 350–400 in diameter. Suckers 98–112 in diameter, unarmed. Evaginated rostellum 290 long and 126 in diameter at apex. Rostellum provided with 10 hooks, 32 long. Strobila 196 wide immediately posterior to base of scolex. Genital pores unilateral, dextral. Genital ducts passing dorsal to poral longitudinal excretory canals and ventral to aporal canals. Cirrus finely spinose. Cirrus sac measuring 256 by 22, extending beyond aporal excretory canals. External seminal

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vesicle sacculate, 128 by 41, lying dorsal to and looping back over cirrus sac. Single testis ovoid to spherical, attaining size of 64 by 60 in mature proglottids; located in middle of segment. Ovary, lobate, 60–80 long, located anterior and ventral to testis, contained within boundaries of ventral excretory canals. Vitelline gland, 35 by 28, situated ventral and posterior to ovary, usually between posterior lobes of latter. Vagina about 5 in diameter, posterior and ventral to cirrus sac. Seminal receptacle prominent. Completely gravid segments not observed. Ventral longitudinal excretory canals about 9 in diameter; dorsal canals 4 in diameter.

Type host: Polysticta stelleri (Pallas).

Type locality: Point Barrow, Alaska.

Habitat: Duodenum.

Type specimen: One slide, USNM No. 49,437, containing the type and paratype specimens, has been deposited in the Helminthological Collection of the U. S. National Museum.

TABLE I.—*List of cestodes identified from eider ducks collected in Alaska and data on prevalence, host numbers, and collection localities*

Host	Steller's eider	Pacific eider	King eider	Spectacled eider
Locality	Point Barrow	Point Barrow Wainwright St. Lawrence Island Amchitka Island	Point Barrow Wainwright St. Lawrence Island	Point Barrow Yukon Delta*
Total number	10	36	32	15
Number males	4	14	17	7
Number females	6	22	15	8
Total number infected	7	21	22	13
Cestodes obtained (In order of frequency of occurrence)	*Hymenolepis sibirica* von Linstow, 1905 *Haploparaxis polystictae* n. sp.	*Lateriporus teres* (Krabbe, 1869) *Hymenolepis microsoma* (Creplin, 1829) *Hymenolepis fallax* (Krabbe, 1869) *Hymenolepis fausti* (Tseng Shen, 1932) *Fimbriarioides intermedia* (Fuhrmann, 1913) *Hymenolepis tenuirostris* (Rudolphi, 1819) *Hymenolepis arctica* n. sp.	*L. teres* *F. intermedia* *H. fallax* *H. microsoma* *H. arctica* n. sp.	*H. arctica* n. sp. *L. teres* *Hymenolepis sacciperum* Mayhew, 1925 *Hymenolepis yukonensis* Schiller, 1954 *Diorchis flavescens* Krefft, 1871 *Schistocephalus solidus* (Müller, 1776)

* This material was considered in another paper (Schiller, 1954).

Discussion: According to recent literature (Rausch, 1951; Schiller, 1951a, 1951b; Flores-Barroeta, 1953), 5 of the 27 known species of the genus *Haploparaxis* Clerc, 1903, occurring in birds are characterized by their possession of spinose cirri and rostellar hooks of a shape considered "typical" for the genus (i.e., having a short handle relative to the length of the guard and blade). These are: *H. clerci* Yamaguti, 1935; *H. scolopacis* Yamaguti, 1935; *H. xemae* Schiller, 1951; *H. galli* Rausch, 1951; and *H. rissae* Schiller, 1951. Of these, *H. polystictae* n. sp. most closely resembles *H. scolopacis* (parasitic in CHARADRIIFORMES) on the basis of rostellar hook size. However, the new species differs from *H. scolopacis* in the shape of the rostellar hooks, in length of the strobila (about 12 mm. for *H. polystictae* n. sp., compared with 270 mm. for *H. scolopacis*), in the shape and extent of the ovary (lobate in *H. polystictae* n. sp.; transversely elongate, extending to both excretory canals in *H. scolopacis*), and in the relative length of the cirrus sac (extending beyond aporal excretory canals in *H. polystictae* n. sp., but usually not reaching mid-line of proglottid in *H. scolopacis*).

The position of the genital ducts relative to the longitudinal excretory canals (genital ducts passing dorsally to poral longitudinal excretory canals and ventral to aporal canals) in this species may be unique for the genus *Haploparaxis;* this

arrangement has not been seen in any other members of the genus examined by the writer, and apparently has not been reported in the literature.

Steller's eider is apparently capable of sustaining heavy cestode infections without ill effect. A young bird of a brood which had just attained ability to fly was collected by Dr. Robert Rausch south of Pt. Barrow. This duck was extremely heavily parasitized with *H. polystictae* n. sp. Transverse histological sections[1] of the intestine disclosed that the scolices of these cestodes were deeply embedded in the submucosa of the duodenum (Fig. 17). Despite the severity of the infection, and the depth to which the cestodes had penetrated in the intestinal wall, nothing was observed to indicate that the well-being of the host was adversely affected.

Hymenolepis arctica n. sp.

(Figs. 4–7)

(Measurements in microns, unless otherwise designated.)

Diagnosis: Length of strobila about 60–80 mm.; maximum width 2 mm., attained near posterior end of strobila. Scolex 148 by 176. Suckers 48 in diameter, directed forward, weakly muscled and unarmed. Evaginated rostellum 136 long and 40 in maximum diameter at apex. Rostellum provided with single row of 10 hooks, 15 long. Strobila 84 in diameter at narrowest place immediately posterior to base of scolex. Genital primordia appear about 4 mm. posterior to scolex. Genital pores unilateral and dextral. Genital ducts pass dorsal to excretory canals. Cirrus sac averages 448 in length and 35 in diameter External seminal vesicle pyriform, 150 by 64. Cirrus slender and aspinose. Sacculus accessorius and cirrus stylet absent. Testes 3, arranged in straight line and appearing trypanosome-like in shape during early stages of development, becoming irregularly lobed, attaining size of 198 by 96 in mature proglottids. Testes lying in same plane between excretory vessels. Ovary deeply lobed with numerous small finger-like projections. Vitelline gland irregular in shape, measuring 70 by 35. Ovary and vitelline gland situated slightly aporal to median line of proglottid and posterior to aporal testis. Vagina lying posterior and ventral to cirrus sac. Vagina forming characteristic loop before reaching genital atrium. Voluminous seminal receptacle in mature proglottids prominent, measuring 336 long and about 140 in maximum diameter. Uterus extending transversely across median line of proglottid as a slender irregular tube, passing dorsal to excretory canals and developing by enlargement, filling entire proglottid when gravid. Completely developed eggs not observed. Ventral longitudinal excretory canals measuring 22 in diameter, dorsal canals, 5.

Type host: Somateria spectabilis (L.); also in *Somateria mollissima v-nigra* Gray, and *Arctonetta fischeri* (Brandt).

Type locality: Point Barrow, Alaska.

Habitat: Duodenum.

Type specimen: One slide, USNM No. 49,438, containing the type and paratype specimens, has been deposited in the Helminthological Collection of the U. S. National Museum.

Discussion: Comparison of *H. arctica* n. sp. with other members of this genus, characterized by the presence of 10 rostellar hooks of a similar size and comparable shape, and lacking cirrus spines, a cirrus stylet, and a sacculus accessorius, revealed that the new species bears some resemblance to the following: *H. microcephala* (Rudolphi, 1819); *H. recurvirostroides* Meggitt, 1927; *H. haldemani* Schiller, 1951; *H. microcirrosa* (Mayhew, 1925); and *H. fusa* (Krabbe, 1869). The significant morphological differences which serve to separate these species from *H. arctica* n. sp. are enumerated below:

H. microcephala (parasitic in ARDEIFORMES) has a strobila of much greater length (300 mm.) and is characterized by the unusual disposition of the testes (poral and aporal testes lying without the boundaries of the excretory canals).

H. recurvirostroides (parasitic in CHARADRIIFORMES) has a considerably shorter

[1] The histological materials used in this study were prepared by Reggie V. Rausch, of this laboratory.

strobila (30 mm.) and a smaller cirrus sac (not extending beyond poral excretory canal.)

H. haldemani (parasitic in LARIFORMES) has testes of subspherical to oval shape which are disposed in a triangular pattern. One testis, located poral to ovary, lies in a plane dorsal to ovary and aporal testes.

H. microcirrosa (parasitic in PASSERIFORMES) has testes of subspherical shape, the cirrus sac extends only slightly beyond poral excretory canals, and the seminal receptacle is not characteristically prominent.

H. fusa (parasitic in LARIFORMES) attains a much greater length (90 mm.), the testes are subspherical in shape, and the cirrus sac extends in a straight line to the middle of the segment.

The trypanosome-like shape of the testes (Fig. 6) in pre-mature segments of *H. arctica* n. sp. was consistent in the 41 specimens examined in this study. This feature is regarded as a useful aid in recognizing the species.

Hymenolepis fallax (Krabbe, 1869)
(Figs. 13–14)

The original specimens of *H. fallax* were collected from the Pacific eider in Greenland in 1867. Although this species can be differentiated from the other members of this genus by the number, size, and shape of its rostellar hooks, the rest of the anatomy is poorly known. Therefore, a redescription of this species is presented herein, based on material from both the Pacific and king eiders collected at Point Barrow, Alaska.

(Measurements in microns, unless otherwise designated.)

Diagnosis: Length of strobila about 280 mm.; maximum width 4 mm. Scolex 196 in in diameter. Suckers 80 in diameter, unarmed. Evaginated rostellum 182 long and 73 in diameter at apex. Rostellum provided with single row of 28 to 30 hooks, 9 to 11 long. Strobila 126 wide immediately posterior to base of scolex. Genital pores unilateral and dextral. Cirrus densely spinose. Cirrus sac measuring 518 by 84 and extending to poral excretory canals. Ovoid to sacculate external seminal vesicle lying ventral to cirrus sac and averaging 510 by 280 in mature proglottids. Testes 3, elongate to irregular in shape, about 350 by 154, and arranged in straight line with one poral and two aporal to ovary. Poral testis flattened antero-posteriorly in post-mature proglottids. Ovary deeply lobed and located in middle of proglottid. Vitelline gland irregularly shaped, postovarian. Vagina usually directly ventral to cirrus sac. Sacculate seminal receptacle very prominent. Uterus extending transversely across median line of proglottid and developing by enlargement, becoming sacculate when gravid, passing dorsal to and extending beyond excretory canals, nearly reaching margins of proglottid. Ventral longitudinal excretory canals measuring 112 in diameter; dorsal canals 14 in diameter. Completely developed eggs not observed.

Type host: Somateria mollissima (L.); also recorded by Krabbe (1869) from *Aythya marila* (L.) in the Faeroe Islands.

Type locality: Greenland.

One slide, USNM No. 49,440, containing an entire specimen from *Somateria spectabilis* (L.), collected at Point Barrow, Alaska, has been deposited in the Helminthological Collection of the U. S. National Museum.

Variabilty: The specimens of *H. fallax* from both the Pacific and king eiders show a considerable degree of variation in number and distribution of the testes. A single strobila, selected at random, contained 698 proglottids. In this cestode the number of testes ranged from 1 to 4, and 7 different testicular arrangements were seen. The proglottids with unusual testicular distribution were spaced at irregular intervals throughout the strobila. The combination occurring most fre-

quently was that of 1 testis poral, and 2 aporal, to the ovary. The 6 additional variants seen, and the frequency of their occurrence were as follows: 2 testes poral and 1 aporal to ovary, 8 times; 2 testes poral and 2 aporal to ovary, 3 times; 1 testis poral and 3 aporal to ovary, 3 times; 1 testis poral and 1 aporal to ovary, 3 times; no testis poral and one aporal to ovary, 5 times; no testis poral and 2 aporal to ovary, 3 times. Comparable variations in number and arrangement of the testes have been recorded for other species in the genus *Hymenolepis* (see Mayhew, 1925; Voge, 1952; Schiller, 1952).

The poral testis in late mature proglottids of *H. fallax* is extremely distorted as compared with that of the aporal testes (Fig. 14). The generally flattened, elongate shape of the poral testis, in this case, appears to be due to the pressure of the distended seminal receptacles in adjacent segments. These already occupy nearly all of the area within this part of the proglottid long before the testes attain maximum size. This appears to be another example in which antero-posterior development of the testis is restricted by crowding of other organs, as was observed by the writer (1952) to be the case in some specimens of *Hymenolepis horrida* (von Linstow, 1905).

Hymenolepis sibirica (von Linstow, 1905)

(Figs. 8–10)

Von Linstow (1905) described *Diorchis sibirica* on the basis of fragmentary cestodes from the king eider. The original material was restudied by Fuhrmann (1908), who transferred the species to the genus *Hymenolepis*. Insofar as the writer can determine, the collection of *H. sibirica* has not been reported since the original description.

Cestodes obtained from Steller's eider during the present work are identical with *H. sibirica* insofar as can be ascertained from von Linstow's description. Von Linstow did not describe the scolex, but comparisons of the following morphological characters were possible: strobila length and width; cirrus sac size and shape; cirrus spination; seminal vesicle size and shape; extent of seminal receptacle; and egg size. Of these, cirrus sac size and shape, cirrus spination, and egg size are relatively consistent characters and are regarded as having taxonomic value.

The fact that the hosts of von Linstow's and the writer's cestodes are closely related also seems to support the assignment of the present material to *H. sibirica*.

The following redescription of *H. sibirica* is based on the specimens from Steller's eider, collected at Point Barrow, Alaska:

(Measurements in microns, unless otherwise designated.)

Diagnosis: Length of strobila about 300 mm.; maximum width 3 mm. Scolex 96 by 214. Suckers elongate, 67 by 32, weakly muscled and unarmed. Evaginated rostellum about 196 long and 21 in diameter at apex. Rostellum provided with a single row of 10 hooks, 25 in length. Hooks confined within partially retracted rostellum in specimens available for study. Strobila about 48 in diameter at narrowest place immediately posterior to scolex. Genital primordia appear about 4.2 mm. posterior to scolex. Genital pores unilateral and dextral. Genital ducts passing dorsal to ventral longitudinal excretory canals. Cirrus sac averaging 420 by 57. External seminal vesicle sacculate, 192 by 96. Cirrus thick and densely covered with heavy spines. Sacculus accessorius and cirrus stylet absent. Testes 3, ovoid, arranged in straight line, 2 poral and 1 aporal to ovary and vitelline gland. Poral testes averaging 224 by 140 in mature proglottids. Aporal testis slightly larger with an average size of 280 by 154. Testes all in same plane. Aporal and poral testes sometimes overlapping ventral longitudinal excretory canals, but never occurring completely outside boundaries of latter. Ovary, usually

consisting of 3 main lobes, situated slightly aporal to mid-line of proglottid. Transverse measurements of ovary in mature proglottid averaging 420. Vitelline gland irregular in shape, 126 by 84, situated slightly ventral and just posterior to mid-line of ovary. Vagina thin-walled; margins appearing crenated in optical section (possibly an artifact resulting from fixation). Ovoid to sacculate seminal receptacle measuring about 210 to 140 in post-mature proglottids. Uterus extending transversely across median line of proglottid as an irregular tube and developing by enlargement, filling entire proglottid when gravid. Eggs averaging 52 by 32. Hooks of embryo 7 long. Ventral longitudinal excretory canals measuring 70 in diameter; dorsal canals, 14.

Type host: Somateria spectabilis (L.)

Type locality: Taymyr Peninsula.

Three slides, USNM No. 49,436, containing a single entire specimen from *Polysticta stelleri* (Pallas), collected at Point Barrow, Alaska, have been deposited in the Helminthological Collection of the U. S. National Museum.

Considerable variation in the number of testes was noted in all of the writer's specimens of *H. sibirica.* As an example, in a strobila comprised of 861 segments, 147 contained only 2 testes. In view of this situation, and the poor condition of the original material (according to Fuhrmann), it is understandable why von Linstow described this species as a member of the genus *Diorchis.*

Fimbriarioides intermedia (Fuhrmann, 1913)

(Figs. 11, 12, 18)

Fuhrmann (1913) originally assigned this species to the genus *Fimbriaria* Frölich, 1802. Later (1932) he proposed the genus *Fimbriarioides,* to which *F. intermedia* was transferred as the type species. In a revision of the subfamily FIMBRIARIINAE, Webster (1943) emended the genus *Fimbriarioides* to include a new species, *F. haematopodis,* which he described from the black oystercatcher, *Haematopus bachmani* Audubon. In differentiating the members of this genus, he concluded that the ovoid shape of the testes and the number of longitudinal muscle bundles (140–150) served to distinguish *F. intermedia,* despite the fact that its scolex was unknown.

Cestodes belonging to the genus *Fimbriarioides,* complete with scolices, were obtained from both the Pacific and king eiders in Alaska. The shape of the testes was ovoid in these specimens and transverse sections of the strobila (cut at 10 μ) showed the presence of 158 longitudinal muscle bundles (Fig. 18). The rest of the morphological characters were in agreement with the original description of *F. intermedia.* According to this Alaskan material, the scolex of *F. intermedia* is described as follows: Length, about 160 μ; width, 144 μ. Suckers circular, weakly muscled, unarmed, and average about 57 μ. Rostellum about 108 μ in length and about 35 μ in diameter at apex. Rostellum provided with 10 hooks, 22 μ in length. Strobila 80 μ wide immediately posterior to base of scolex.

One slide, USNM No. 49,441, containing an entire specimen, has been deposited in the Helminthological Collection of the U. S. National Musuem.

Lateriporus teres (Krabbe, 1869)

(Figs. 15–16)

Cestodes from the Pacific eider, collected in Greenland, and from the old squaw duck, *Clangula hyemalis* (L.), taken in Finnmark, Norway, were described as *Taenia teres* by Krabbe (1869). Fuhrmann (1907) also studied cestodes occurring in Pacific eider in Greenland, which he concluded were conspecific with those de-

scribed by Krabbe; however, after reexamination of Krabbe's original material, he proposed the genus *Lateriporus* and designated *L. teres* as the type species. Some additional details of the morphology of this species were included in Fuhrmann's report.

Skrjabin (1926) also recorded *L. teres* from the Pacific eider, obtained during the Russian Arctic Expedition of 1912–1914. Although the locality is not indicated in Skrjabin's report, these ducks probably were taken in the vicinity of Franz-Josef Land, where many birds were collected by this expedition.

Specimens of *L. teres* from Pacific, king and spectacled eiders in this study agree, for the most part, with the original description. The only difference appears to be in the spinose condition of the cirrus in the Alaskan material. Since both Krabbe's and Fuhrmann's descriptions were based on fragmentary specimens, it is quite probable that the cirrus spines had been lost; therefore, this difference is not regarded as significant. The more important morphological characters of *L. teres* from the Alaskan birds are given in Table 2. The shape of the rostellar hooks

TABLE 2.—*A comparison of the measurements of* Lateriporus teres
(Measurements in microns, unless otherwise designated.)

	L. teres (Krabbe, 1869)	*"L. geographicus"* Cooper, 1921	*L. teres* (Alaskan material)
Length	420 mm.	172 mm.	up to 405 mm.
Width	3 mm.	2 mm.	3 mm.
Number of rostellar hooks	12–16	Unknown	16
Size of rostellar hooks	150–170	Unknown	154
Size of cirrus sac	290 by 20	240 by 70–90	280 by 70
Seminal vesicle	Absent	Not mentioned (absent in fig.)	Absent
Cirrus spines	Absent	Present	Present
Number of testes and size	30 (100 by 50)	15–20 (80–120)	15–30 (80–126)
Size of seminal receptacle	110	150 by 120	160
Shape of ovary	Deeply lobed	Deeply lobed	Deeply lobed
Egg	Round	125–145 in diameter	Capsule: up to 132 Inner shell: 74 by 67
Size of onchosphere	?	55	54–60
Size of hooks of onchosphere	21–23	?	24

is illustrated in Figure 16, and a drawing of the egg of this species is shown in Figure 15.

Synonymy: Cooper (1921) described *Lateriporus geographicus* on the basis of cestodes occurring in the Pacific eider, collected during the Canadian Arctic Expedition of 1913–1918. Alcohol-preserved specimens, designated as *"Lateriporus geographicus* Cooper (paratype)" were made available to the writer through the kindness of Dr. E. L. Bousfield, Division of Zoology, National Museum of Canada, Ottawa. These cestodes apparently had, at some time, undergone severe desiccation and, although parts of strobilae were stained and mounted, they were unsuitable for study. According to Cooper's original description of *L. geographicus,* the rostellar hooks were not observed, but the rest of the morphological characters appear to be within the limits of variation seen in *L. teres* (see Table 2) and it is concluded that these species are conspecific.

Three slides, USNM No. 49,439, containing 1 specimen of *L. teres* from *Somateria spectabilis* (L.), collected at Point Barrow, Alaska, has been deposited in the Helminthological Collection of the U. S. National Museum.

LITERATURE CITED

COOPER, A. R. 1921 Trematodes and cestodes of the Canadian Arctic Expedition, 1913–18. Rep. Canad. Arctic Expedition 1913–18, 9, parts G-H: Trematoda and Cestoda. pp. 3–27.

FLORES-BARROETA, L. 1953 Céstodos de vertebrados. I. Ciencia. **13:** 31–36.

FUHRMANN, O. 1907 Bekannte und neue Arten und Genera von Vogeltänien. Centralbl. Bakteriol. Orig. **45:** 516–536.

———1908 Die Cestoden der Vögel. Zool. Jahrb. Suppl. **10:** 232 pp.

———1913 Nordische Vogelcestoden aus dem Museum von Göteborg. Göteborgs K. Vetensk.-o Vitterhets-Samh. Handl. 4 f. (1911–12) **14–15,** Medd. Göteborgs Mus. Zoolog. Avd. (1), 41 pp.

———1932 Les ténias des oiseaux. Mém. Univ. Neuchâtel. **8:** 1–381.

KRABBE, H. 1869 Bidrag til Kundskab om Fuglenes Baendelorme. K. Danske. Vidensk. Selsk. Skr., Naturv. og Math. Afd. **8:** 249–363.

MAYHEW, R. L. 1925 Studies on the avian species of the cestode family Hymenolepididae. Ill. Biol. Monogr. **10** (1): 1–125.

RAUSCH, R. 1951 Studien an der Helminthenfauna von Alaska. IV. *Haploparaxis galli* n. sp., ein Cestode aus dem Schneehuhn, *Lagopus rupestris* (Gmelin). Zeitschr. Parasitenk. **15:** 1–3.

SCHILLER, E. L. 1951a Studies on the helminth fauna of Alaska. I. Two new cestodes from Sabine's gull (*Xema sabini*). J. Parasit. **37:** 266–272.

———1951b Studies on the helminth fauna of Alaska. VIII. Some cestode parasites of the Pacific kittiwake (*Rissa tridactyla* Ridgway) with the description of *Haploparaxis rissae* n. sp. Proc. Helminth. Soc. Washington, **18:** 122–125.

———1952 Studies on the helminth fauna of Alaska. X. Morphological variation in *Hymenolepis horrida* (von Linstow, 1901) (Cestoda: Hymenolepididae). J. Parasit. **38:** 554–568.

———1954 Studies on the helminth fauna of Alaska. XVIII. Cestode parasites in young Anseriformes on the Yukon delta nesting grounds. Trans. Amer. Micr. Soc. **73:** 194–201.

SKRJABIN, K. I. 1926 Izuchenie gel'mintologicheskoĭ kollektsii sobrannoĭ ekspeditsieĭ G. Îa. Sedova k Severnomu Poliûsu v 1912–1914 godu. Trudy Gosudarstu. Inst. Eksper. Vet. **4:** 114–121.

VOGE, M. 1952 Variation in some unarmed Hymenolepididae (Cestoda) from rodents. Univ. Cal. Pub. Zool. **57:** 1–52.

VON LINSTOW, O. 1905 Helminthen der Russischen Polar-expedition 1900–1903. Mém. Acad. Sc. St. Pétersb. Cl. Phys.-Math., 8 s., **18:** 17 pp.

WEBSTER, J. D. 1943 A revision of the Fimbriariinae (Cestoda: Hymenolepididae). Tr. Am. Micr. Soc. **62:** 390–397.

EXPLANATION OF PLATES

PLATE I

FIG. 1. Scolex of *Haploparaxis polystictae* n. sp.

FIG. 2. Rostellar hook of *H. polystictae* n. sp.

FIG. 3. Mature proglottid of *H. polystictae* n. sp. (ventral view).

FIG. 4. Scolex of *Hymenolepis arctica* n. sp.

FIG. 5. Rostellar hook of *H. arctica* n. sp.

FIG. 6. Pre-mature proglottid of *H. arctica* n. sp. showing characteristic shape of testes during early stage of development.

FIG. 7. Mature proglottid of *H. arctica* n. sp. (ventral view).

FIG. 8. Scolex of *Hymenolepis sibirica.*

FIG. 9. Rostellar hook of *H. sibirica.*

FIG. 10. Mature proglottid of *H. sibirica* (dorsal view).

FIG. 11. Scolex of *Fimbriarioides intermedia.*

FIG. 12. Rostellar hook of *F. intermedia.*

FIG. 13. Rostellar hook of *Hymenolepis fallax.*

FIG. 14. Mature proglottids of *H. fallax* (dorsal view).

FIG. 15. Egg of *Lateriporus teres.*

FIG. 16. Rostellar hook of *L. teres.*

PLATE I

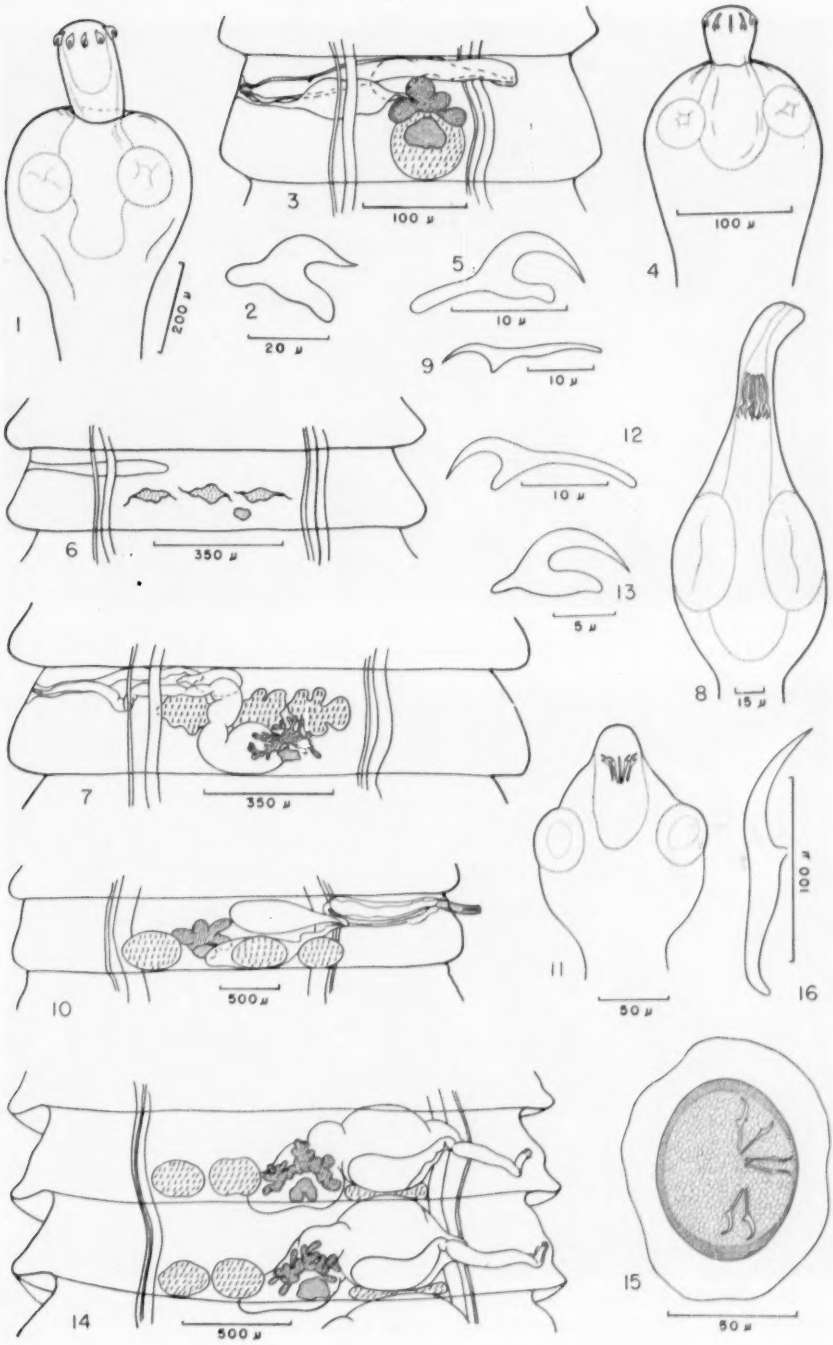
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PLATE II

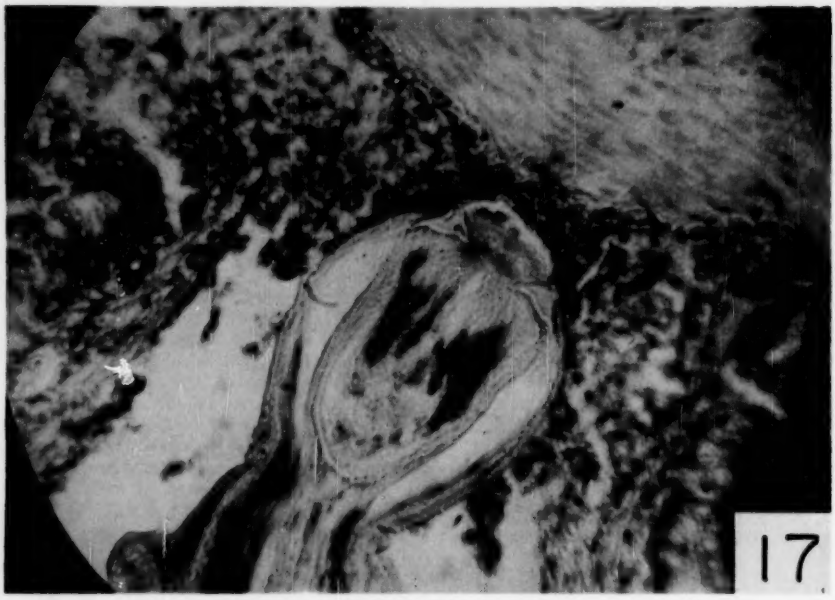

FIG. 17. Photomicrograph of histological section of intestine of Steller's eider showing embedded rostellum of *Haploparaxis polystictae* n. sp. (×2000).

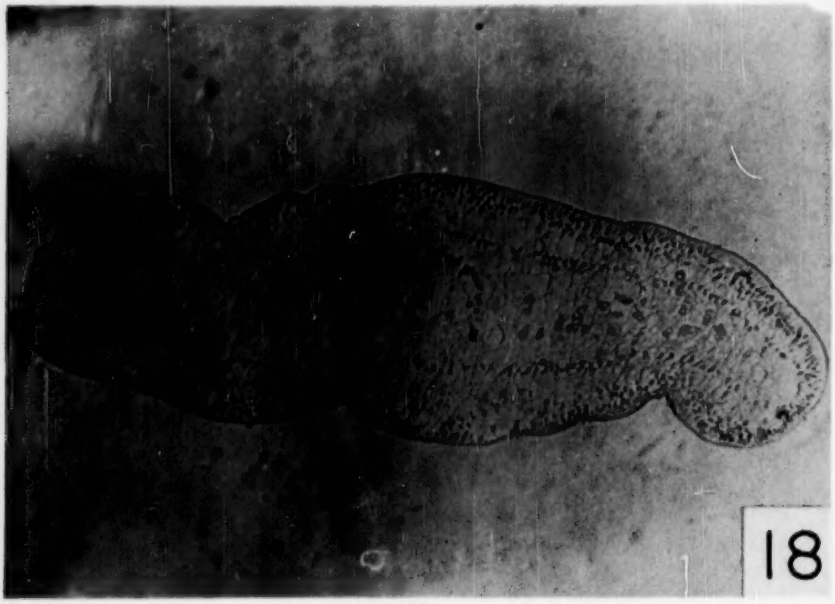

FIG. 18. Photomicrograph of transverse histological section of *Fimbriarioides intermedia* showing number and position of longitudinal muscle bundles (×50).

NEMATOPSIS OSTREARUM AND *N. PRYTHERCHI* (EUGREGARININA: POROSPORIDAE) WITH SPECIAL REFERENCE TO THE HOST–PARASITE RELATIONS

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INTRODUCTION

Nematopsis ostrearum (EUGREGARININA: POROSPORIDAE) is the first known American member of a family of gregarines unique in having alternation of hosts. This parasite, having vegetative stages in certain mud crabs and spore stages in the American oyster, was discovered by Prytherch and described by him in detail (1938, 1940) after he had carried out the entire life cycle experimentally. Prytherch performed other experiments (unpublished) and made extensive field observations which led him (1940) to the belief that *Nematopsis* is an important factor in extensive mortalities occurring periodically along the Gulf and Atlantic coasts. The same author (1940) gave a complete historical account of the POROSPORIDAE, composed of the genera *Porospora* and *Nematopsis*.

The possible relationship between *Nematopsis* and oyster mortalities has been a disputed question and a matter of serious concern to many people with scientific or economic interests in oysters. Published experimental data seem to be few or lacking, although many field studies have been made and a few of them, notably that by Landau and Galtsoff (1951), have been published.

Although the present study was undertaken primarily to obtain experimental data relative to the possible lethal effect of *Nematopsis* on oysters and other matters of host-parasite relation, the opportunity was taken to test the opinion formed by the senior author in preliminary observations that the two types of spores mentioned by Prytherch (1940) represent two species. The senior author, at the same time, extended the observations made by Prytherch (1938, 1940) on morphology and life cycle.

Nematopsis ostrearum Prytherch has already been separated into *N. ostrearum*, emended, and a new species, *N. prytherchi*, by Sprague (1949) in an abstract anticipating a series of papers dealing with these species. In this paper experiments are reported which concern primarily the host-parasite relations, but which also provide part of the basis on which the two species of *Nematopsis* were separated. A later paper will deal more fully with morphology and life cycle of the parasites.

During June, 1948, through January, 1949, three infection experiments were performed at Louisiana State University Marine Laboratory, Grand Isle, La., to obtain, in addition to other data, information on host-parasite relations. These

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were a part of an extensive investigation of factors in oyster mortality conducted by Texas A and M Research Foundation. The first of the three experiments was performed with two main purposes, (1) to test various decapods as possible definitive hosts of species of *Nematopsis* represented by different types of spores in the American oyster, *Crassostrea virginica* (Gmelin), and (2) to determine whether extremely heavy experimental infections with those spores could cause mortality of oysters. Accordingly, the first experiment is reported here in two parts. The other two experiments, more limited in scope, are presented with the latter phase of the first one and treated as second and third mortality experiments.

MATERIALS AND METHODS

Crabs and shrimp were collected from various localities in the vicinity of Grand Isle. All the *Eurytium limosum* and most of the *Panopeus herbstii* were taken from burrows among the mangrove roots on the bank of Bayou Rigaud near the Humble Company docks on Grand Isle. Large specimens, which proved more suitable for the experiments than small ones, were found there in abundance in the summer of 1948. Probably most of the *Panopeus* were *P. herbstii forma obesa.* Most of the specimens of *Eurypanopeus depressus,* a small species, were taken from natural oyster reefs in Mudworm Bayou, Grand Terre Island. *Petrolisthes armatus,* a still smaller species, was found in abundance on shell reefs at an oyster camp near Sugar House Bend in Barataria Bay. *Menippe mercenaria,* the largest of the crabs employed, was found in burrows among oyster shells, under stones, and in crevices among the debris found in certain localities in the intertidal zone. Shrimp were collected anywhere in Barataria Bay by trawling.

Excepting in a part of the experiment involving *Eurytium limosum,* no attempt was made to rid the decapods of intestinal gregarines before using them. Numerous observations had convinced the authors that gregarines present at a given time would complete their development and be eliminated in less than a week at laboratory temperature (about 28°–30° C.); it was presumed, therefore, that any which might be observed as long as a week after the experiment started would be experimentally acquired. Chelae were removed from the larger crabs to prevent their causing damage to the shells of the experimental oysters. The removal was usually accomplished by crushing the chelae with pliers and thereby supplying a stimulus for autotomy. This method proved very satisfactory, whereas merely breaking off the appendages seemed to be accompanied by excessive hemorrhage and often death of the crabs.

Nematopsis cysts (host cells containing spores) in mantle, muscle and heart tissues were counted by a method employed in unpublished studies by Drs. J. H. Roberts and H. W. Harry. This involved treating the tissues with 10% NaOH solution and examining them in a microscope fitted with a Whipple micrometer. In each such examination the cysts in 10 fields, each being 1 mm. square and selected at random, were counted and the average number of cysts per sq. mm. was calculated. The oysters employed contained originally in the margins of the mantles approximately 1.3 *Nematopsis* cysts per sq. mm., this being the equivalent of about 3.6 times as many spores (Wang and Sprague, unpublished studies) or 4.7 spores per sq. mm. No method of counting the spores in the gills, where they are unevenly distributed in dense clusters, has been devised.

Running water was passed into the aquaria through aerating nozzles of a type devised by Mr. L. Boswell who was occupied with other phases of the oyster mortality investigation. The parts of each nozzle were a 4 inch length of 8 mm. rubber tubing with 4 holes punched in radial arrangement near the middle, the glass portion of a medicine dropper and a glass T-tube approximately 8 mm. in diameter. The side arm of the T-tube was inserted into one end of the perforated rubber tube and the narrow portion of the medicine dropper was inserted into the other end. Then the broad end of the medicine dropper was inserted into the water supply line. When properly adjusted these devices caused excellent mixing of air and water flowing through them into the aquaria. The rate of water flow through each aerator and into each aquarium was approximately 1900–2000 ml./min. Although the oxygen content of the water in the aquaria was not tested, it was assumed that the continuous rapid flow of aerated water kept the oxygen level sufficiently high to supply the needs of the experimental animals.

Almost every day throughout the summer of 1948 the meats of approximately a half bushel (before shucking) of oysters were fed to the decapods, this amount of food being about all the decapods would consume. These oysters were collected primarily from Lirette's beds in Bay Saint Elaine and natural reefs in Sugar House Bayou, Grand Terre Island, since occasional sampling indicated that oysters from both sources contained large numbers of the 3 types of *Nematopsis* spores mentioned by Sprague (1949).

I. TESTING DECAPODS AS POSSIBLE VECTORS OF *Nematopsis* IN OYSTERS

Penaeus aztecus Ives

Shrimp from Bay des Islettes were collected on June 8 and 3 of them were examined for the presence of intestinal gregarines. These 3 shrimp, like every one of hundreds examined by us soon after collecting, harbored gregarines resembling *Nematopsis*. On the same day, 50 of the shrimp and 30 oysters from the source already indicated were placed in each of 2 aquaria. The shrimp were fed daily on heavily infected oyster meats and 33 of them were examined between 7 and 29 days after the first feeding. No gregarines, excepting sporozoites which had just emerged from ingested spores, were found. Fifteen of the experimental oysters were examined and the incidence of *Nematopsis* noted. The intensity of infection, indicated in Table 1, showed no increase throughout the experiment but remained similar to that in the original samples and similar to that in the controls. Although many control oysters were examined from time to time, it seems superfluous to tabulate the results. Table 1 will, therefore, be referred to hereafter as an accurate indication of the incidence of *Nematopsis* in similar oysters before they were exposed to other decapods and in control oysters throughout.

Although these and separate observations showed that the sporozoites readily emerged in the mid-gut of shrimp from *Nematopsis* spores ingested with oyster meats, no later stages of the same parasites were ever observed in these decapods. Furthermore, the shrimp lost the gregarines occurring naturally in them and apparently transmitted no parasites to the experimental oysters. These results point clearly to the conclusion that the gregarines occurring naturally in shrimp and the *Nematopsis* spores found in oysters are different species. The gregarine of

TABLE 1.—*Incidence of* Nematopsis, *attributable to previously acquired infection, in oysters exposed to shrimp in the laboratory*

Date examined	No. of cysts/sq. mm. in organs indicated			
	Margin of mantle	Heart	Adductor muscle	Gill
June 11	3.0	0.0	0.7	Spores observed
" "	0.3	0.0	0.1	No spores observed
" "	3.6	0.0	0.0	" " "
" "	0.3	0.0	0.0	" " "
" "	0.2	0.0	1.3	" " "
" "	0.5	0.0	0.0	" " "
" 24	0.3	0.0	0.1	Spores observed
" "	2.4	0.4	0.4	No spores observed
" "	1.9	0.0	0.0	Spores observed
" "	30.0	0.2	1.4	No spores observed
" 25	0.8	0.0	0.0	Spores observed
" "	0.7	0.0	0.0	No spores observed
" "	4.2	0.1	0.5	" " "
" "	0.3	0.0	0.1	" " "
" "	0.4	0.0	0.6	" " "

shrimp, observed many years ago by Prytherch (personal communication) was recently described by Sprague (1954) as a new species of *Nematopsis*.

Petrolisthes armatus (Gibbes)

Two hundred crabs of this species were collected on June 4, 1948, at a locality already mentioned. Six of them were examined during that and the next day and no gregarines were found. On June 7 and almost daily thereafter the remaining crabs were fed infected oyster meats. Six of the crabs were then examined on July 6 and 8 on July 28. Still, no gregarines were found. Oysters exposed to the crabs for about 2 months showed no increase in intensity of *Nematopsis* infection. It must be concluded that the data provide no reason to suppose that *P. armatus* is one of the decapodan hosts of species of *Nematopsis* known in oysters.

Panopeus herbstii Milne Edwards

At the beginning of the experiment 69 small crabs of this species were placed with 30 oysters in each of 2 aquaria. On June 18 10 large specimens were added to each lot. Four crabs were examined over a period of 1 to 3 weeks after the first day of feeding upon infected oyster meats (June 9) and 3 were found to harbor gregarines. It is presumed that these gregarines developed from *Nematopsis* spores ingested under experimental conditions.

The intensity of *Nematopsis* infection in the exposed oysters was, at the beginning of the experiment, similar to that shown in Table 1. After 3 weeks or more of exposure to the crabs, 26 of 27 oysters examined contained enormous numbers of developing and mature spores in the mantles (Table 2). The spores

TABLE 2.—*Incidence of* Nematopsis *in oysters exposed to* Panopeus herbstii *for various intervals June 9 to late August, 1948*

Date of examination	No. examined	No. with obviously great increase of spores in mantle	No. with appreciable increase of spores in gills	No. with little or no increase of spores
June 11–28	9	9	0	0
" 29–July 15	7	6	0	1
July 22–Aug. 26	16	16	0	0
Sept. 28*	4	4	0	0

* Some of the oysters were examined approximately a month after exposure to the infected crabs was terminated.

were mostly situated in a narrow band near to, and parallel with, the margin of the mantle. In one instance, not an unusual case, it was estimated that the concentration of cysts in that region was approximately 1000 (the equivalent of 3600 spores) per sq. mm. Never were natural infections seen with more than about one-third that intensity. Since the spores were present in such great numbers and since it was often very difficult even to see the very slightly refractive immature ones, it was not feasible to try to count them. Counts were unnecessary for comparison with the numbers of cysts in the control oysters, however, because the enormously greater numbers of spores, especially immature ones, in the experimental oysters was obvious and very striking. Even a few immature spores appeared in the gills but they, like the ones in the mantle, muscle and heart, were all of a single type, the type which is most common in the mantle.

The spores attributable to experimental infection when apparently matured were always of one type, being broadly rounded at the ends and measuring approximately 10 by 14–15 microns. They were probably identical, in spite of slight discrepancies in measurement, with spores which Prytherch (1940) described as being 11–12 by 16 microns and which he designated as immature spores of *Nematopsis ostrearum*. Pryterch, who first produced these spores experimentally by exposing oysters to experimentally infected *Panopeus herbstii* and *Eurypanopeus depresseus*, saw also larger spores, 11 by 20 microns, which he considered to be the final or mature stages of the same species. Since, in the present studies, only the smaller of these 2 types of spores developed in oysters exposed to *Panopeus* (the larger, as indicated by data presented later, developed only in oysters exposed to Menippe), it is believed that the larger spores observed by Prytherch during the course of his experiments were already present in his experimental oysters. It is also believed, therefore, that he inadvertently included 2 species under the name of *Nematopsis ostrearum*. Accordingly, the larger spores which Prytherch accidentally included in *N. ostrearum* have been removed to a new species and *N. ostrearum* has been reserved for that species which he described in detail.

Eurypanopeus depressus (Smith)

One hundred crabs of this species were placed in each of 2 aquaria with 30 experimental oysters from Chene Fleure and fed infected oyster meats from June 8 to the latter part of August. Eight of 15 crabs examined between June 17 and August 21 harbored gregarines, probably experimentally acquired. The incidence of *Nematopsis* in these crabs during the experiment was somewhat erratic, for reasons unknown, and the numbers were few. Possibly the size of the species was a factor, for *Eurypanopeus* is a small crab and incapable of ingesting large quantities of food. Perhaps, therefore, these crabs often ingested few or no *Nematopsis* spores with the oyster tissues.

The oysters exposed to the crabs were presumed to have had at first essentially the same frequency of *Nematopsis* spores as those listed in Table 1. When they were examined after several weeks of exposure to the crabs many showed an obviously greater number of spores in the mantle but not in the gills (Table 3).

Some of the spores produced experimentally in the oyster mantles appeared somewhat smaller than those usually seen in instances of natural infection and tended to be pointed at 1 end. They closely resembled the *Nematopsis* spores

TABLE 3.—*Incidence of* Nematopsis *in oysters exposed to* Eurypanopeus depressus *for various intervals from June 8 to late August, 1948*

Date of examination	No. examined	No. with obviously great increase of spores in mantle	No. with appreciable increase of spores in gills	No. with little or no increase of spores
June 11	4	0	0	4
" 25	4	1	0	3
July 6–Aug. 6	5	2	0	3
Aug. 9–Sept.* 25	40	40	0	0

* Some of the oysters were examined approximately a month after exposure to the infected crabs was terminated.

which are common in *Modiolus demissus* occurring in Grand Isle. It is futile to speculate on the significance of these unusual spores without further study. The great majority of the spores, on the contrary, were indistinguishable from those transmitted by *Panopeus*. Pending evidence to the contrary, the authors must tentatively agree with Prytherch (1938, 1940) that *Eurypanopeus depressus* is one of the normal decapodan hosts of *Nematopsis ostrearum*. Further, the evidence indicates that this crab species does not transmit the large spores, *N. prytherchi*, which are characteristically seen in oyster gills.

Eurytium limosum (Say)

On July 15, 1948, 215 large crabs were collected from among the mangrove roots along the bank of Bayou Rigaud near the Humble Company plant on Grand Isle. Twenty-three of them (10.7%) proved to be *Eurytium limosum* and the rest *Panopeus herbstii*. Since the former species was found in such relative abundance and since, according to Rathbun (1930), it is "related to *Panopeus* and its allies," it was also employed in the infection experiment to determine whether it might harbor *Nematopsis* and transmit this parasite to oysters.

Unfortunately, none of the crabs was examined before being fed infected oyster meats. It is not known, therefore, whether they harbored gregarines naturally. The crabs were all placed in an aquarium with 12 oysters, presumed to have *Nematopsis* counts similar to those listed in Table 1, and similar oysters in another aquarium without crabs were employed as controls. The crabs were fed *Nematopsis*-infected oysters daily and examination of crabs and oysters was made according to the schedule given in Table 4. As the data show, all the crabs examined contained

TABLE 4.—*Experimental infection of* Eurytium limosum *and oysters with* Nematopsis ostrearum

No. of days after feeding began, July 17	Crabs			Oysters		
	No. examined	Gregarines present		No. examined	Spores in	
		trophic	cysts		mantle	gill
1	1	Many	0	..	..	..
2	1	"	9	..	..	..
3	2	Many, few	4, ca. 80	..	..	..
5	..	..	..	1	No increase	No increase
6	2	Few, none	6, ca. 200	..	..	..
11	1	Many	ca. 50	1	No increase	No increase
13	..	..	..	1	" "	" "
18	..	..	..	1	Many young	" "
20	..	..	..	6	" "	" "
20	..	..	..	1	No increase	" "
20	..	..	..	6 (controls)	No increase	" "
23	..	..	..	1	Many young and mature	" "

many gregarines. The trophic stages in the midgut and the gamontocysts (a term adopted from Filipponi, 1949, for the older "gametocysts," since these structures are encysted gamonts rather than encysted gametes) attached to the lining of the rectum near the anus could not be distinguished from those occurring in *Panopeus herbstii*. The gymnospores (which are very significant in characterizing species and which will be described in a later paper) of the gregarines in *Eurytium* appeared identical with those in *Panopeus*. Furthermore, the *Nematopsis* spores which developed in the mantles of oysters exposed to the crabs appeared to be those of *Nematopsis ostrearum*.

Although it is not known whether the crabs employed in the experiment harbored gregarines in the beginning, it is believed (because of time and temperature factors involved and because gregarines were observed in all stages of development) that most of those observed were acquired in the laboratory. This belief was supported by another phase of the experiment carried out as follows: When it seemed evident that *Nematopsis* was developing in *Eurytium* in the laboratory the remaining crabs were starved for a week and 2 of them then examined were found negative. The others were fed daily upon infected oyster meats and further examinations were made. The crabs, after feeding, were found to harbor gregarines as shown in Table 5. Again, the morphological features of the parasite (sporadins,

TABLE 5.—*Occurrence in* Nematopsis *in* Eurytium limosum *starved for a week and then fed infected oysters*

Date	No. of crabs examined	Gregarines present	
		In midgut	In rectum
Aug. 27 (after week of starvation)	2	0	0
" 19 (first feeding)	..	..	..
" 20	1	Many large	0
" 22	1	" "	4 young cysts
" 28	1	Many, all stages	Ca. 50 cysts, all stages

gamontocysts, gymnospores, resistant spores) appeared identical with those of *Nematopsis ostrearum*. There seems to be little doubt, therefore, that infection of *Eurytium limosum* was experimentally produced in the laboratory and that the infection was further transmitted to oysters. Since the entire life cycle of the parasite was completed under laboratory conditions, another decapod host of *Nematopsis ostrearum* was added to the list of those already made known by Prytherch. Confirmation, however, by means of more refined experiments is desired.

Menippe mercenaria (Say)

These crabs were fed *Nematopsis*-infected oyster meats on June 9 and almost daily thereafter during the experiment. Ten of them were examined (between 1 week and 2 months after feeding began) and all contained gregarines which were probably acquired in the laboratory. All the major developmental stages, sporozoites to mature gamontocysts with gymnospores, were seen in great numbers. The majority of the stages observed were not noticeably different from corresponding stages of *Nematopsis ostrearum* seen in *Panopeus herbstii, Eurytium limosum* and *Eurypanopeus depressus*. The gymnospores seen in *Menippe* (in this and other feeding experiments as well as in natural infections) were, however, always conspicuously larger than those seen in other crabs. This fact strongly supports others

which point to the conclusion that *Menippe* harbors a species of *Nematopsis* other than *N. ostrearum*.

Oysters from Chene Fleure, presumed to have *Nematopsis* counts similar to those given in Table 1, were exposed to the crabs for varying lengths of time (beginning June 11) up to more than 2 months. During June (beginning June 18), July and August 36 of the oysters were examined microscopically. The gills of these oysters, without exception, contained large numbers of *Nematopsis* spores, mostly immature and usually of two types which are commonly seen in naturally infected gills and which differ only in size (Sprague, 1949). The great majority were, however, of the larger type. In many instances the entire gill appeared to be literally saturated with spores, each microscopic field containing apparently many thousands. The intensity of the infection was incomparably greater than any seen in nature. Appreciable, but not very great, numbers of similar spores were often observed in mantle, muscle and heart tissues; but no increase in numbers of the type usually observed in these organs was noted. The distribution of the spores developing in the mantle was also noteworthy. Unlike the usual spores in mantle tissue, they were not concentrated in a band parallel with the margin but were scattered, sometimes in clusters, throughout the mantle tissues.

Since oyster meats fed to *Menippe* contained (in addition to the usual 2 forms in the gills) typical spores of *Nematopsis ostrearum* but similar spores did not develop in the oysters exposed to this crab, it is presumed that *N. ostrearum* was not capable of developing in *Menippe*. The spores which were transmitted are, furthermore, presumed to represent one or more different species.

From the experiments and observations already recorded here 4 significant types of evidence have emerged which, taken together, strongly support the view that the large *Nematopsis* spores having special affinity for oyster gills and the smaller ones having special affinity for the mantle (which Prytherch designated as respectively mature and immature stages of *N. ostrearum*) represent two distinct species. These types of evidence are: (1) morphological differences in resistant spores; (2) morphological differences in gymnospores; (3) differences in decapod host specificity; (4) marked differences in distribution of spores in the molluscan host. It has already been stated that the name *Nematopsis ostrearum* probably should be reserved for all the stages which Prytherch described exclusive of that which he designated as the mature spore. In a preliminary note Sprague (1949) has already given the latter stage a new name, *N. prytherchi*, in honor of Dr. Herbert F. Prytherch who first reported it. He tentatively assigned also to the new species the smaller but otherwise similar spores transmitted to oyster gills by *Menippe*.

Although the authors believe that the evidence presented above justifies the separation of species, they recognize a flaw in the experiments which permits another possible interpretation of the results. Namely, it was necessary, because the only oysters available for feeding the crabs had mixed infections, to feed more than one kind of a spore at the same time. Then, when morphologically different parasites developed in the different crabs and these gave rise to morphologically and physiologically different spores in oysters, it was assumed that there was a selective affinity between a particular crab species and a particular type of infective spore. An alternative assumption is that the different types of spores developed indiffer-

ently in the various crab species. This would suggest that the differences observed in the parasites could possibly have been differences within a single species caused by passage through different crab hosts. The conclusions reached here, therefore, require confirmation by further experimentation employing more suitable materials.

II. TESTING THE CAPACITY OF *Nematopsis* TO KILL OYSTERS

Certain aspects of the host-parasite relation, particularly those incidental to the ascertaining of decapodan hosts of *Nematopsis ostrearum* and *N. prytherchi*, have already been considered. The rest of this paper deals more fully with the interaction between host and parasite but especially with the effect of the parasite on the molluscan host. The latter was studied by means of a series of infection experiments in which an attempt was made to introduce into the oysters lethal numbers of *Nematopsis* spores. The statement of materials and methods already given is generally applicable to these experiments but essential details are added below.

First mortality experiment: As already stated, the first mortality experiment was one phase of that experiment reported above and was designed also to test various decapods as possible primary hosts of species of *Nematopsis* under consideration.

Oysters exposed to crabs in this experiment were from Landry's beds in Chene Fleure and contained relatively few *Nematopsis* spores, the numbers being as shown in Table 1. They were of 3 lots which had been collected at different times and/or handled differently before they were used in the experiment. The experiment was set up between the inclusive dates June 6 and June 11, 1948. Twenty-five or 30 oysters (depending on size) were placed in each of several small (18" by 24" by 10") wooden aquaria with running sea water. Two of the aquaria were employed for oysters exposed to each of the crab species and 1 for each lot of control oysters. Chelae were removed, when they were large, from the crabs and these animals were daily fed large quantities of *Nematopsis*-infected oysters. The experimental oysters were observed daily and deaths noted.

TABLE 6.—*Results of first mortality experiment*

Crabs employed	Species of *Nemotopsis* introduced into oysters	Oysters			
		Lot no.	No. employed	Mortality	
				Number	Percent
[illegible]	None (control)	1	21	4	19.0
N. nippe	*N. prytherchi*	1	44	40	90.9
None	None (control)	2	27	19	70.4
Panopeus	*N. ostrearum*	2	56	33	58.9
None	None (control)	3	28	10	35.7
Eurypanopeus	*N. ostrearum*	3	54	16	29.6

Since early mortalities may have been attributable to the changed environment, oysters which died before June 18 were simply replaced and mortality records were kept beginning that date. A few oysters were sacrificed for microscopic examination during the course of the experiment and these were deducted from the original numbers before mortality percentages were calculated. Most of those which died were examined microscopically. Those exposed to the crabs, as already indicated, generally contained unusually large numbers of *Nematopsis* spores but the controls did not. The mortality data from June 18 to August 30, 1948, are shown in Table 6.

Second mortality experiment: Since the first experiment gave data of questionable significance, another was performed with the hope that some of the complicating factors might not enter into it. That experiment was likewise complicated by unknown mortality factors and the results are of doubtful value. It is, therefore, presented only briefly here.

Oysters employed were the largest obtainable and were collected from 2 sources with respectively low and high incidence of *Nematopsis*. The object of the experiment was to test the possible effect of heavy experimental infection of *Nematopsis* on large oysters which had not been previously heavily infected and on others which already naturally harbored relatively large numbers of spores. The first source was Bassa Bassa, in Barataria Bay, where the natural incidence of *Nematopsis* was relatively slight. The other oysters, from Cheramie's beds in Barataria Bay, harbored numerous *Nematopsis* spores.

The oysters were distributed in large wooden aquaria with aerated running sea water. Some were exposed to *Nematopsis* infected crabs and others without crabs were considered as controls. The experiment was begun on July 6 and terminated on September 20, 1948. As shown in Table 7, most of the control oysters

TABLE 7.—*Results of second mortality experiment*

Crabs employed	Species of *Nematopsis* introduced into oysters	Oysters			
		Source	Number employed	Mortality	
				No.	Percent
None	None (control)	Bassa Bassa	64	57	89.1
Menippe	*N. prytherchi*	" "	49	49	100.0
Panopeus	*N. ostrearum*	" "	50	50	100.0
None	None (control)	Cheramie	80	73	91.2
Menippe	*N. prytherchi*	"	80	80	100.0
Panopeus	*N. ostrearum*	"	80	80	100.0

died. Apparently, mortality factors other than *Nematopsis* were operative. The *Nematopsis* parasites possibly contributed to the deaths of the oysters exposed to the crabs, however, because they all died and most of them showed a great increase in the number of *Nematopsis* spores.

Third mortality experiment: Twenty-five oysters from Chene Fleure, as nearly uniform in size as possible and acclimated to laboratory conditions for 1 week, were placed in each of 2 small aquaria with running sea water aerated as already described. With them in each tank were placed 25 *Menippe mercenaria* with chelae removed. The crabs were fairly uniform in size, varying within a range of 12 mm. Almost daily the crabs were fed *Nematopsis*-infected oysters obtained from Lirette's beds in Bay St. Elaine and Staufflet's beds in Bayou Bas Bleu. Two other aquaria, for controls, were set up in identical manner except that the crabs were previously starved for a week to rid them of *Nematopsis* and were thereafter fed only small fish and shrimp. Every attempt was made to maintain similar conditions in the experimental and control aquaria excepting the 1 factor, *Nematopsis prytherchi*, in the former. Two similar experimental and 2 control aquaria were set up with *Panopeus herbstii* to test the possible effect of *Nematopsis ostrearum* on the oysters.

Each day, with few exceptions, an inventory was made of the crabs and oysters to note mortalities, and the water temperature and rate of flow was recorded. Most of the oysters were examined microscopically after they died and the incidence of

Nematopsis was noted. The experiment was carried out over a period of 2.5 months (Nov. 1, 1948, Jan. 15, 1949). The mortality data are shown in Table 8.

It will be noted that, for both species of the parasites, there was somewhat higher mortality among the experimentally infected oysters than among the controls. These differences, although not large, support the slight evidence obtained in the other experiments suggesting that heavy experimental infections of *Nematopsis* may have contributed to the oyster mortalities. It is noteworthy, also, that there was a slightly higher mortality among the infected crabs than among the controls. It is possible the parasites have some deleterious effect on the decapodan hosts.

DISCUSSION

The mortality experiments: It is unfortunate that healthy and *Nematopsis*-free oysters were not available for the experiments, for the use of such material might have yielded more significant results. There was no alternative to superimposing an experimental infection upon a natural one and hoping that the results would be susceptible of interpretation. Although the experimental infections were incomparably more intense than natural ones, their effects cannot be decided with confidence because the effect of spores already present is unknown. Furthermore, it is quite

TABLE 8.—*Results of third mortality experiment*

Species of *Nematopsis* introduced	Crabs				Oysters		
	Kind	No. employed*	Mortality		No. employed	Mortality	
			No.	Percent		No.	Percent
None (control)	*Menippe*	54	4	7.4	50	11	22.0
N. prytherchi	"	83	33	38.4	50	19	38.0
None (control)	*Panopeus*	59	9	15.3	50	9	18.0
N. ostrearum	"	61	11	18.0	50	24	48.0

* The number of crabs in each aquarium was kept constant (25) by replacing those which died. The numbers given in this table are combined totals for two aquaria.

probable that the experiments were complicated by highly lethal factors other than *Nematopsis*. In view of the indecisive results, Prytherch's (1940) view that *Nematopsis* is highly lethal to the molluscan host has neither been confirmed nor refuted. The data may show a trend which suggests that massive doses of *Nematopsis* spores are deleterious to oysters. Yet, the fact that some of the oysters containing extremely great numbers of spores survived the experiments leaves the authors unconvinced that the much smaller numbers of spores occurring naturally contribute very significantly to mortality of the host.

Distribution of the spores in the oyster

Spores of *Nematopsis prytherchi* may occur in blood cells of almost any of the organs of the molluscan host. They have been seen in appreciable numbers in labial palps, mantle, heart, adductor muscle, and among the liver tubules. They are by far most abundant, however, in the blood vessels of the gills, particularly in the proximal region. In bulk samples of fresh gill tissues they appear as independent spores packed together in large masses, unlike those of *N. ostrearum* which are contained within highly granular and very conspicuous host cells ("cysts") somewhat scattered in the involved tissues. Sections show plainly, however, that spores of *N. prytherchi* are also contained within cysts that have limiting mem-

branes which may be the principle remnants of greatly hypertrophied and otherwise modified phagocytes. Whether the cysts of the two species are derived from the same type of host cell is not certain. If they are, the host cells respond differently to the 2 parasites and produce in the one instance cysts which are conspicuous and in the other cysts which are invisible in bulk samples of tissue. Cysts of *N. prytherchi* seem to adhere to the walls of the blood vessels and to each other. In sections they constitute a continuous reticulum containing the spores in its interstices. Possibly, there is a specific agglutinating reaction which is the basis of the special affinity of those cysts for the gills. Whatever the reason, the cysts accumulate on the walls of the blood vessels of the gills and cause the lumina to become narrowed or completely occluded. This would seem to be a serious impediment to the circulation of blood and might interfere with the proper functioning of the gills, a possibility already suggested by Prytherch (1940).

N. ostrearum and only 1 previously described species (*Nematopsis* sp. Schneider, 1892) have special affinity for the mantle, all others being most concentrated in the gills. This statement does not take into account spores reported by Sprague (1954) in mantle and gills of *Modiolus demissus* and others in mantle of *Ensis minor* because that report was based on casual observations only. The cysts of *Nematopsis ostrearum* are concentrated in a narrow band, roughly 2 mm. wide, near to, and parallel with, the margin of the mantle. They do not occur in clumps, as do those of *N. prytherchi*, but are rather uniformly distributed within the band. The reason for the peculiar concentration of cysts was not determined by us. Cysts in the mantle are not limited to that part already mentioned but may occur in any region, even in tentacles, in much smaller numbers. They also occur, as numerous investigators have observed, in considerable numbers in adductor muscle, heart, gill, labial palps, and perhaps any other organ. The results of counts of cysts in mantle, muscle and heart of several oysters suggested little or no correspondence between numbers of cysts in the different organs.

From the foregoing discussion it is apparent that both species of *Nematopsis* under consideration may occur in considerable numbers in many organs of the molluscan host. The statement that one has special affinity for mantle and the other for gill may, therefore, be strictly accurate when only these 2 organs are compared.

Molluscan-host specificity

Previous investigators have generally held the view that a given species of *Nematopsis* is capable of developing in a considerable number of molluscan species. Thus, Léger and Duboscq (1925), Hatt (1927, 1931) and Prytherch (1938, 1940) each listed many mollusc species supposed to be hosts respectively of *N. portunidarum*, *N. legeri* and *N. ostrearum*. Those lists seem to have been compiled largely on the basis of casual observations of somewhat similar spores occurring naturally in the various molluscs. One investigator, Hatt (1931), believed that he had demonstrated experimentally that *N. legeri*, a normal parasite of *Mytilus galloprovincialis*, could inhabit a mollusc other than its usual host. The present writers arrive at just the opposite conclusion from Hatt's experiment, since Hatt was able to obtain only very slight development of the parasite in several weeks in the unnatural host, *M. minimus*. It is suggested here that more careful comparison of the

Nematopsis spores occurring naturally in different molluscs, combined with critical experiments, might result in the recognition of several other distinct species of *Nematopsis* and lead to a modification of the generally held opinion that each of these parasites is capable of developing to the mature spore stage in a great variety of molluscs.

Effect of parasite on the decapodan host

During the course of the experiments described above numerous crabs were subjected to repeated heavy infections of *Nematopsis* over a long period of time. Occasional mortalities were attributable to mechanical injury or, when the water flow stopped for a few hours, to asphyxiation. In only one experiment was a record of the crab mortalities kept. In that instance, as already pointed out, there was somewhat greater mortality among the experimental animals than among the controls (Table 8). Although the data are very limited, the gregarine parasites possibly contributed to these mortalities.

Distribution of the parasites in the crabs

Vegetative stages of POROSPORIDAE, as well as other gut-inhabiting gregarines, are characteristically situated in the anterior portion of the mid-gut. Young trophozoites in this family are attached to epithelial cells by means of a spherical epimerite. Later trophic stages are free in the lumen. Gamontocysts are cemented to the chitinous lining of the rectum. The location in the rectum is a characteristic of the species, some species (*Porospora gigantea*, *P. nephropis*, *Nematopsis prytherchi* and *N. penaeus*) being distributed along the entire length of this organ, and others (*N. ostrearum*, *N. legeri*, *N. maraisi* and *N. portunidarum*) being concentrated at the extreme posterior end near the anus. The particular distribution of the parasites in the rectum probably depends on particular environmental conditions corresponding to the specific requirements of the parasite. It may be that those species situated at the anus require for their development some contact with the sea water. More complete dependence upon the sea water environment for gamontocyst development is illustrated by *Carcinoecetes conformis*, a crab-inhibiting gregarine possibly related to the POROSPORIDAE and studied by Ball (1838). In this species the gamontocysts are usually found attached to the telson and various abdominal appendages of the host.

Decapodan-host specificity

The findings with respect to host specificity of the parasites under consideration have already been presented and need not be repeated at this time. Suffice it to say that they are in agreement with those of previous investigators who have found that the spores will germinate indifferently in many species of decapods but the sporozoites can survive and continue their development only in one or few host species. For most of the POROSPORIDAE, including *Nematopsis prytherchi*, a single decapodan host is known for each species. *N. ostrearum*, however, appears to have at least 3 decapodan hosts (of as many genera) and *N. portunidarum*, according to Léger and Duboscq (1913), has 2 or possibly 3. It is quite probable that *N. penaeus*, as pointed out by Sprague (1954), inhabits more than one species of *Penaeus*.

General remarks on gregarines as parasites

The cephaline gregarines generally are not noted for being highly detrimental to their hosts, the best known effect being damage done to the epithelial cells to which the young trophozoites are attached in the intestine of the host. This effect, whatever it may be, is directly related to the number of parasites present, since each parasite attacks a single host cell. The number present at a given time is limited by two important factors inherent in the process of reproduction. In the first place, the EUGREGARININA (to which category *Nematopsis* belongs) are peculiar in having no asexual reproduction. The vast majority of the parasitic PROTOZOA (as well as pathogenic bacteria) undergo some type (or types) of asexual reproduction (binary fission, multiple fission, budding, plasmotomy, etc.) to produce large numbers of new individuals like the parents which are capable of spreading the infection within the body of the host. These unique PROTOZOA, the EUGREGARININA, reproduce only by sporogony (sexual reproduction) and the products leave the body of the host to perish or enter again the same or another host. Thus, these parasites do not increase numerically within the host by reproduction. In this respect they are comparable to many helminth parasites rather than to other PROTOZOA. Unlike the helminths, however, the adult gregarines themselves perish in the process of sexual reproduction; this is the second limiting factor, mentioned above, in the numbers of parasites in the host. Therefore, the reproductive process in these gregarines not only fails to increase but actually decreases the numbers of individuals present in the host. This process causes complete and rapid (the time being a function of the temperature) elimination of the gregarines present at a given time and the infection, if it is maintained, must be replenished by new invaders. The number of parasites (and possibly their effect), therefore, fluctuates and the severity of their effect may depend very largely on whether there is repeated and frequent acquisition of new parasites. Acquisition of great numbers possibly results in damage to the host other than the destruction of epithelial cells as mentioned above. It is probable that the sheer bulk of numbers may damage the intestine mechanically or interfere with the proper function. There may also be a toxic effect of the parasites, this being a debatable subject discussed by Watson (1916).

The above remarks apply either to certain gregarines which have only one host or to those stages of the POROSPORIDAE occurring in the definitive host (the one in which sexual reproduction occurs, the decapodan host of POROSPORIDAE). In *Nematopsis* the possible effect of the parasite on the intermediate (molluscan) host is of particular interest because of the economic importance of oysters. Here, again, there is no known multiplication within the host but each individual parasite (spore) infecting the oyster is acquired as an original invader. Any effect of the parasite on the host must, therefore, be caused by these original invaders; there can be no effect (as in most SPOROZOA and in pathogenic bacteria) attributable to multiplication of the parasite within the oyster. Spores of *Nematopsis*, unlike the vegetative stages, may, according to observations by Landau and Galtsoff (1951) as well as by Prytherch (1940) and the present writers, be cumulative. This suggests the possibility of a cumulative effect on the host. It also seems quite possible that such an effect may accompany the progressive mechanical obstruction of the blood vessels of the organs involved. It is also conceivable, as Prytherch (per-

sonal communication) has suggested, that toxic end products of metabolism might be liberated into the host tissues by the developing sporoblasts. The authors have no data pertinent to the latter hypothesis.

SUMMARY AND CONCLUSION

Experiments are here reported which provided a partial basis on which *Nematopsis ostrearum* as described by Prytherch (1938, 1940) has already been separated into *N. ostrearum* Prytherch, emended, and *N. prytherchi* Sprague, 1949.

N. prytherchi includes only one stage described by Prytherch, viz. large spores which are more common by far in oyster gill than in mantle and which Prytherch mistakenly regarded as final developmental stages of spores belonging to the species described by him in detail. Vegetative stages of *N. prytherchi* occur in the crab *Menippe mercenaria*, the only known primary host of this parasite.

Nematopsis ostrearum, emended, includes smaller spores, which are rare in oyster gills but common in mantle and other organs, and the vegetative stages which Prytherch described in the crabs *Panopeus herbstii* and *Eurypanopeus depressus*. The experiments herein reported led to the conclusion that *Eurytium limosum* is also host to this gregarine.

Experiments failed to produce any evidence that either the crab *Petrolisthes armatus* or the shrimp *Penaeus aztecus* is host to the species of *Nematopsis* found in oysters, although this shrimp harbors naturally a gregarine resembling *Nematopis*.

Extremely large numbers of spores of each species were introduced into oysters in an attempt to kill the hosts. The results were complicated by unknown mortality factors and were neither consistent nor decisive. There was a slight trend toward greater mortality among oysters into which *Nematopsis* was introduced than among controls. It is tentatively concluded that the experimental infections, incomparably greater than any known in nature, may have contributed to the mortality. It is felt, however, that the experimental data lend no support to the view that the numbers of spores which have been observed to occur naturally in oysters are very significant factors in mortality.

Very limited data suggest that *Nematopsis* may, in some instances, be significantly deleterious to the decapodan host.

Increase of numbers of *Nematopsis* parasites in the decapodan or, so far as is known, in the molluscan host is never by multiplicative reproduction within the host but only by new invasion.

From the decapodan host the parasites tend to be rapidly and completely eliminated by their reproductive process. If there is replenishment it is by new invasion and the severity of any deleterious effect on the host may depend largely on whether there is frequent acquisition of large numbers of new parasites.

In the molluscan host the number of parasites seems to be cumulative. This suggests the possibility that any deleterious effect on the host may be related to numbers of parasites acquired over a period of time.

The value of the experimental data might have been enhanced had *Nematopsis*-free oysters been available for use and had it been possible to control extraneous mortality factors.

REFERENCES CITED

BALL, G. H. 1938 The life history of *Carcinoecetes hesperus*, n. gen., n. sp., a gregarine parasite of the striped shore crab *Pachygrapsus crassipes*, with observations on related forms. Arch. Protistenk. **90**: 299–319.

FILIPPONI, A. 1949 Gregarine policistides parassite di *Laemostenus alegerinus* con considerazioni sulla nomenclatura nelle gregarine. Riv. Parassitol. **10** (4): 245–263.

HATT, P. 1927 Spores de Porospora (Nematopsis) chez les gastéropodes. Compt. Rend. Soc. Biol., Paris, **96**: 90–91.

———1931 L'évolution des porosporides chez les mollusques. Arch. Zool. Expér. et Gen. **72**: 341–415.

LANDAU, H. AND GALTSOFF, P. S. 1951 Distribution of Nematopsis infection on the oyster grounds of the Chesapeake Bay and in other waters of the Atlantic and Gulf States. Texas J. Sci. **3**: 115–130.

LÉGER, L. ET DUBOSCQ, O. 1913 Le cycle évolutif de *Porospora portunidarum* Frenzel. Compt. Rend. Acad. Sci., Paris **156**: 1932–1934.

———1925 Les porosporides et leur évolution. Trav. Stat. Zool. Wimereaux **9**: 126–139.

PRYTHERCH, H. F. 1938. Life-cycle of a sporozoan parasite of the oyster. Science **88**: 451–452.

———1940 The life cycle and morphology of *Nematopsis ostrearum*, sp. nov. a gregarine parasite of the mud crab and oyster. J. Morphol. **66**: 39–65.

RATHBUN, M. J. 1930 The cancroid crabs of America of the families Euryalidae, Portunidae, Atelecyclidae, Cancridae and Xanthidae. Bull. 152 U. S. Nat. Mus. pp. 1–607.

SCHNEIDER, A. 1892 Signalement d'un nouveau sporozoaire. Tablettes Zool. Poitiers **2**: 209–210.

SPRAGUE, V. 1949 Species of Nematopsis in *Ostrea virginica*. J. Parasit. **35** (6 Sect. 2): 42.

———1954 Protozoa. In: Gulf of Mexico—its Origin, Waters and Marine Life. Paul S. Galtsoff, Ed. U. S. Fish and Wildlife Service, Fish. Bull. **55** (89): 243–256.

WATSON, M. E. 1916 Studies on gregarines. Including descriptions of twenty-one new species and a synopsis of the eugregarine records from the Myriapoda, Coleoptera and Orthoptera of the world. Illinois Biol. Monogr. **2** (3): 213–468 (258 pp).

SEXUAL ANOMALIES IN THE MALE *SCHISTOSOMA JAPONICUM* (FORMOSAN STRAIN)[1]

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Separation of the sexes in the family SCHISTOSOMATIDAE was considered to be complete until Vogel (1947) discovered hermaphroditism in an African strain of *Schistosoma mansoni*. He described hermaphroditic worms having the build and organs characteristic of the male but provided in addition with an ovary situated posterior to the testes and anterior to the junction of the intestinal ceca. In individual worms, where the female organ was particularly well developed, typical egg cells were seen in the ovary; in some specimens an oviduct and uterus were found but a female genital pore could not be detected. Rudimentary vitellaria were seen in some specimens. No relationship was apparent between the occurrence of vitelline glands and ovarian development. In Vogel's opinion, the transformation of males into hermaphrodites was especially prone to occur in relation to certain species of mammalian hosts such as guinea pigs, hamsters and rabbits. The absence of a female partner seemed to influence the production of a greater proportion of hermaphrodites than when equal or near equal numbers of male and female worms

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were present. Vogel came to the conclusion that this sexual abnormality was confined to *S. mansoni* because under similar experimental conditions no hermaphrodites of *S. japonicum* or *S. haematobium* were found.

Short (1948) recorded finding 13 hermaphrodites among 35 male specimens of the Puerto Rican strain of *S. mansoni* purchased from a commercial supply company. Consequently, no data concerning the mammalian host or the age of the worms were available. In general, Short's observations paralleled those of Vogel except that Short found some worms in which the female components had attained a more advanced stage of development than any observed by Vogel. Several worms exhibited a common genital pore produced by the union of terminal parts of the female and male ducts. Short also confirmed Vogel's observations of the tendency of the male organs to be suppressed in the hermaphrodites.

LaGrange and Scheecqmans (1949) and Buttner (1950) have also reported the occurrence of hermaphrodites in male specimens of *S. mansoni*, presumably of African origin.

Gönnert (1949) reported the occurrence of certain small compact groups of nuclei which he called "resting nuclei" in all male specimens of *S. mansoni* obtained from experimental mice. These nuclei were distributed in clusters between the intestinal ceca from their junction to the genital pore. On the basis of their location alone, these nuclei were interpreted as being rudimentary female sex primordium. Gönnert also described cells located ventral and ventro-lateral to the intestinal ceca which he considered to be rudimentary vitellaria. Considerable variation in the number of testes (0-19) occurred in the male specimens. To us it seems very likely that these anomalies can be attributed to the chemotherapeutic agent, Miracil D, to which the worms had been subjected. Gönnert also reported similar "resting nuclei" in male specimens of *S. japonicum* and *S. haematobium* but he gave neither description nor any information as to the mammalian source of this material.

By finding hermaphroditic female specimens of *Schistosomatium douthitti* Short (1951) has shown that hermaphroditism is not restricted to the male worms of the genus Schistosoma. As in the male Schistosoma, the supernumerary sex organs in the hermaphrodites of *S. douthitti* were situated posterior to the normal sex organ and anterior to the junction of the intestinal ceca. Nearly half of the hermaphroditic female *S. douthitti* observed by Short possessed other abnormalities such as suppression of the vitellaria, absence of eggs in the uterus and in a few worms, ovarian abnormalities. The testicular follicles in the females varied considerably in size and shape; they had no efferent ducts and they were all immature.

Najim (1951) reported finding in an albino mouse a male *S. mansoni*, (Egyptian strain) with a previously undescribed anomaly, namely the presence of supernumerary testes. The supernumerary testes comprised five lobes posterior to the normal set of 9 testicular lobes. No attachments or ducts could be found in connection with the supernumerary testes.

PRESENT FINDINGS

In the course of experimental studies on a Formosan strain of *S. japonicum* we have encountered 3 male worms with sexual anomalies which we believe constitute the first observation of such phenomena in a species other than *S. mansoni*. Our

strain was originally obtained from infected *Oncomelania formosana* supplied by Dr. Eloise B. Cram of the National Institutes of Health, Bethesda, Maryland. Our specimens were recovered from hamsters infected with cercariae emerging from *O. formosana* reared and exposed in this laboratory.

The first anomaly is a male hermaphrodite recovered from a hamster autopsied 59 days after infection to cercariae (Figs. 1 and 2). From this hamster, a total of 44 worms were recovered: 8 sexually mature males in copula with 8 sexually mature females and 28 sexually immature females of smaller size. The abnormal male possesses an ovary situated between the intestinal ceca, 2.7 mm. posterior to the last testicular lobe and 0.9 mm. anterior to the junction of the intestinal ceca. The ovary measures 0.36 mm. in width at the posterior end. The cells forming the anterior portion of the ovary are typical immature ovarian cells, while those in the posterior portion have undergone at least partial development. An oviduct can be seen proceeding from the posterior end of the ovary. Ovarian cells are visible in the area of the fertilization chamber of the oviduct. The oviduct turns and proceeds anteriorad along the side of the ovary to a point 0.18 mm. in front of the anterior margin of the ovary where it enters a well defined oötype. A tube can be seen leading from the anterior portion of the oötype but it soon disappears from view. No evidence of vitellaria or of a vitelline duct could be found. This male worm possesses 7 normal testicular lobes and a seminal vesicle filled with spermatoid cells and spermatozoa.

A second sexual anomaly takes the form of supernumerary testes. The first specimen was recovered from a hamster which was autopsied 60 days after exposure and yielded 34 worms, of which 18 were sexually mature males and 16 mature females. One of the 18 males (Fig. 3) presented 2 supernumerary testicular lobes situated between the intestinal ceca, 0.6 mm. posterior to the normal group of testes. The supernumerary testes are spherical and about 0.12 mm. in diameter. The normal testicular lobes in this specimen are subovate, the smallest of the group measuring 0.13 mm. in long axis. The anterior lobe of the supernumerary testes is provided with a vas efferens which can easily be traced anteriorad because it is filled with spermatids and spermatozoa. No connecting duct can be seen on the posterior supernumerary testes owing to an overlapping of two lobes. The vas deferens of the supernumerary testes appears to dip beneath the most posterior lobe of the normal group of testes and cannot be traced further. A thin-walled vesicle is found between the sixth and seventh testicular lobes of the normal group and extending dorsally. It contains spermatids and spermatozoa and is therefore interpreted to be a seminal vesicle connected to the supernumerary testes by the aforementioned vas deferens. This worm possesses the normal complement of 7 fully developed testicular lobes and a seminal vesicle containing spermatids and spermatozoa in the usual position.

The second worm with a supernumerary testicular lobe was recovered from a hamster exposed 54 days previously. A total of 31 worms were recovered from this animal; 7 were sexually mature males in copula with 7 sexually mature females and 17 were sexually immature females. One of the 7 male worms possessed a single round testicular lobe, 0.6 mm. posterior to the last lobe of the normal group of 7 testes. This worm was mounted in a position that afforded an excellent frontal view of the supernumerary testis; yet no connecting ducts could be seen.

The present finding of hermaphroditism in a male *S. japonicum* from a hamster which harbored an excess of female worms is not entirely in discord with Vogel's findings since he also found hermaphrodites in animals which harbored nearly equal numbers of both sexes. No information was obtained which might help explain the production of supernumerary testes.

After finding these anomalies in specimens of the Formosan strain of *S. japonicum* at least 400 male specimens of the Japanese and Chinese strains of *S. japonicum*, that had accumulated in our laboratory, were re-examined. The worms were derived from unisexual and bisexual infections in various hosts, (mice, hamsters, rats, guinea pigs, rabbits, and a dog) but no further material with anomalies of the sex organs was uncovered.

SUMMARY

Sexual anomalies, in the form of male hermaphrodites and the occurrence of supernumerary testes in male worms of *Schistosoma japonicum* (Formosan strain) are reported. An examination of many more worms of the Chinese and Japanese strains of *S. japonicum* revealed no such anomalies.

REFERENCES CITED

BUTTNER, A. 1950 Curieux cas d'hermaphrodisme chex une souche africaine de *Schistosoma mansoni* (*plathelminthe, trematode*), Compt. rend. des seances de l' Acad. des Sci., Paris **230:** 1420–1422.

LAGRANGE, E. AND SCHEECQMANS, G. 1949 La bilharziose expérimentale du cobaye Compt. Rend. Soc. Biol., Paris **143:** 1396–1399.

GÖNNERT, R. 1946 Über rudimentäre weibliche Geschlectsanlagen bei *Bilharzia mansoni* Männchen. Ztschr. Tropenmed. Parasitol. **1:** 272–279.

NAJIM, A. T. 1951 A male *Schistosoma mansoni* with two sets of testes. J. Parasit. **37:** 545–546.

SHORT, R. B. 1948 Hermaphrodites in a Puerto Rican strain of *Schistosoma mansoni*. J. Parasit. **34:** 240–242.

——— 1951 Hermaphroditic female *Schistosomatium douthitti* (Trematoda: Schistosomatidae). J. Parasit. **37:** 547–555.

VOGEL, H. 1947 Hermaphrodites of *Schistosoma mansoni*. Ann. Trop. Med. Parasitol. **41:** 266–277.

EXPLANATION OF PLATE

All drawings were made with the aid of a cramera lucida from stained totomounts of the worms.

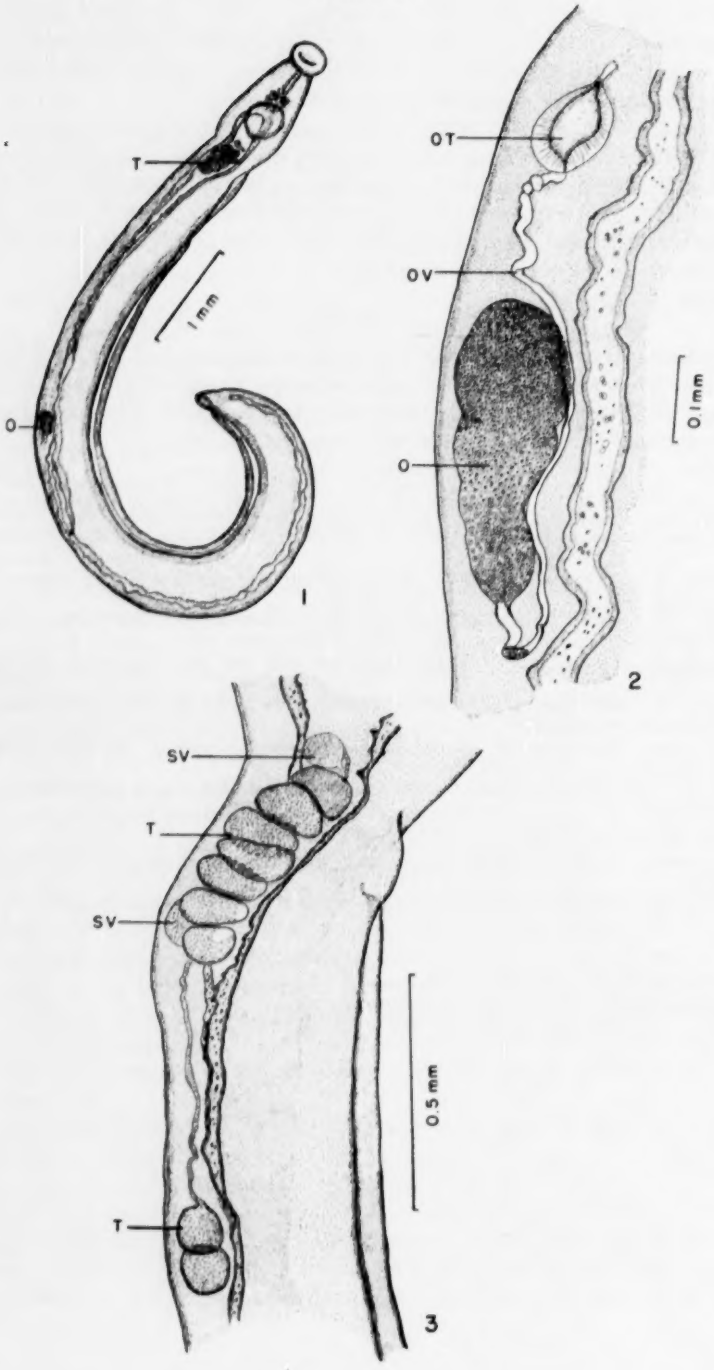
Abbreviations: O-ovary, OD-oviduct, OT-oötype, SV-seminal vesicle, T-testes.

FIG. 1. Male *S. japonicum* with an ovary.

FIG. 2. Female portion of the hermaphroditic male.

FIG. 3. Supernumerary testes in a male *S. japonicum*.

PLATE I



NOTES ON THE REARING OF *ONCOMELANIA* SPP. IN THE LABORATORY

J. H. Sandground and Donald V. Moore*

So complex and obscure are the elements which constitute the environment of snails in nature that maintaining them for experimental purposes over extended periods in temperate zone laboratories is always a precarious task. Despite the overall success enjoyed by this laboratory in continuously maintaining the three human schistosomes for several years, the achievement has been a tenuous one. On more than one occasion thriving colonies of the Oriental species of *Oncomelania* and of the African *Bulinus* have suddenly been decimated by intangible influences making it necessary to reimport snails from their autochthons in order to keep the life cycles going. Two recent innovations which appear to afford improvements over older procedures seem worthy of publication for the guidance of other workers. The first involves the maintenance of *Oncomelania* in a movable habitat small enough to permit low power microscopic observation of the snail and its behavior; the second deals with a new preparation of food in a form that precludes its decay with consequent contamination of the environment.

Of the several molluscan vectors of human schistosomiasis by far the most difficult to breed and maintain in an artificial environment are the various species of *Oncomelania* that transmit *Schistosoma japonicum*. Vogel (1948) described an aquaterrarium which was used successfully by him for over 10 years in maintaining a colony of *Oncomelania hupensis*. The aquaterrarium with minor individual modifications has been used successfully by other laboratories, as well as our own, for various species of these gill breathing, semi-aquatic snails (De Witt, 1952). A rectangular aquarium can easily be converted to an aquaterrarium by building up a bank of sandy loam at one end and creating a shallow ditch or moat inundated with spring water at the other. The morning dew and high humidity of the natural environment which are important for these amphibious snails are simulated in the aquaterrarium by installing the atomizing device described by De Witt (1951). In such aquaterraria the water reservoir, about 2 inches deep, is likely to become fouled if food such as lettuce or cabbage leaves or dried maple leaves accumulate and undergo decay. Under such circumstances the well balanced aquatic environment, which is essential for the young snails, is likely to deteriorate and the young snails are unlikely to survive. In any event the study of the life history and the separate maintenance of the young snails which are used for experimental infections is difficult in a large tank. In order to obtain information such as the number of exposed snails which survive the prepatent period and the number of snails which become infected and shed cercariae it is necessary to keep exposed snails in such a way that they are easily accessible for periodic examinations. To this end we have devised aquaterraria in petri dishes of 10 cm. and 15 cm. diameter. These glass dishes are merely small editions of the large aquaterraria being com-

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* From the Department of Preventive Medicine, New York University, College of Medicine. This investigation was supported by a research grant (G-385) from the National Institutes of Health, Public Health Service.

posed of a sloping sandy loam bank supported by and divided off from a spring water reservoir by a wooden or glass partition. The bank is usually water logged and the atmosphere in the dish is kept humid by the glass lid which covers the dish except at such times as its contents are under observation (Figure 1). When these miniature aquaterraria are made up using the water from a well established biologically balanced large aquaterrarium the associated organisms such as aquatic annelids, protozoa, etc. develop more rapidly and the habitat soon becomes indentical to the one from which the thriving snails had been removed.

So far as is known snails are essentially vegetarian in eating habits. It is customary in most laboratories to feed aquatic snails on cabbage or lettuce leaves either fresh or wilted in hot water to break down the rigid walls of the epidermal cells. For amphibious snails such as the Oncomelanias, fresh leaves have been usefully

FIG. 1. Miniature aquaterrarium showing experimentally exposed *Oncomelania formosana* feeding on food-impregnated paper strips.

supplemented by dried (mature) leaves of the maple tree, which when allowed to steep in the water of a tank seem particularly attractive to the molluscs. In addition some workers have used tropical fish food sprinkled on the ground near the edge of the water (De Witt, 1952). The main objections to this type of diet have been the difficulty and expense of procuring the preferred Roumaine variety of lettuce in winter and the tendency of uneaten and excess of fish food to decay, thus upsetting the delicate biological equilibrium of a successfully established tank. Standen (1951) greatly ameliorated the fresh food problem for rearing *Australorbis glabratus* when he propounded a dried milk, wheat germ and powdered lettuce leaf mixture in sodium alginate. This alginate food was modified and adapted by Moore, Thillet, Carney and Meleney (1953) to utilize ingredients more readily available on the American market. Moore *et al.* (1953) also found this food to be useful for *Bulinus* and *Oncomelania*. This alginate food must be poured in thin layers in order to insure complete transformation of the soluble sodium alginate

into insoluble alginate gel. It is sometimes difficult to judge the proper amount of food to be used especially in aquaria containing only a few young snails; after a few days the uneaten food begins to disintegrate and removal of uneaten food is difficult. We have achieved the objective of producing a thin layer of the food, which is desirable for feeding young snails, and have overcome the objection involved in trying to eliminate decayed leftover food by impregnating filter paper or ordinary paper towelling (such as manufactured by Scott & Co.) with the soluble sodium alginate and then coagulating with calcium chloride solution. The procedure adopted by us is to pour a generous portion of the warm sodium alginate mixture into an 11 by 14 inch developing tray. A sheet of paper towelling is introduced into the mixture and allowed to remain for a few seconds before being drawn

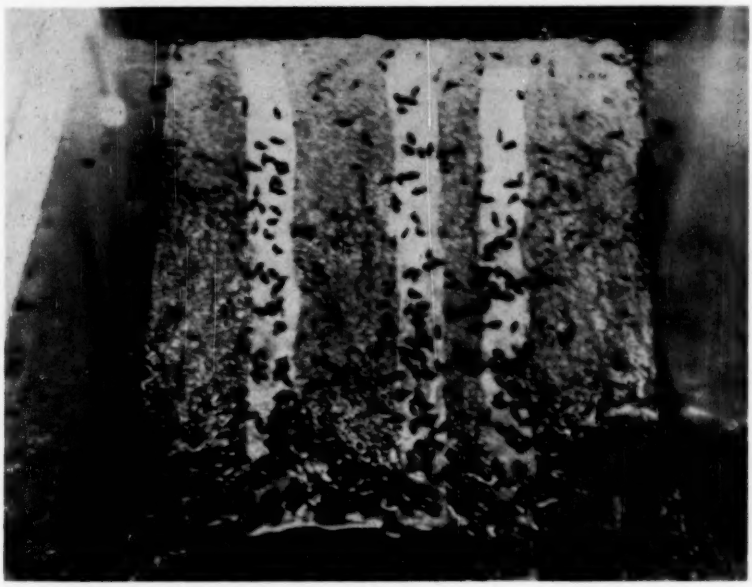

FIG. 2. Food-impregnated paper strips in a large aquaterrarium containing *Oncomelania nosophora*. Twenty-four hours before this picture was taken all snails were removed from the earthen bank and the paper strips laid down.

over the edge of the tray to remove the excess liquid food. The sheet thus impregnated is then transferred to a clean tray and flooded with a 2.5% aqueous solution of calcium chloride. After rinsing in water, the sheet is cut into strips approximately 1 inch wide and stored under water in the 6° C. refrigerator. Strips of food-impregnated paper are offered daily or as frequently as seems necessary.

In feeding *Oncomelania* the paper strip food is set down so that its end dips in the water reservoir while the remainder extends onto the earthen bank of the aquaterrarium. Within a short time snails will have migrated from the four quarters of the tank to the strips of food as illustrated in Figure 2. Strips of food-impregnated paper which have been on the bank for several days may easily be removed to the water reservoir when new strips are added to the bank. The snails which are in the water at the time congregate on the submerged strips and feed

as seen in Figure 3. The paper strip food is a convenient method of presenting food to experimental snails in the aforementioned miniature aquaria. Old strips may be easily removed and replaced with fresh strips thus preventing an accumulation of debris which tends to foul the habitat and upset the balance.

It is commonly assumed that the young forms of *Oncomelania* are more susceptible to infection with *S. japonicum* than are the adult snails. The young stages of *Oncomelania* appear to be strictly aquatic at least for the first weeks of life. Vogel (1948) kept broken pieces of clay flower pots in his aquaterraria and found that he could collect the young of *O. hupensis* from these. Although we have kept broken pieces of clay flower pots in our aquaterraria of *O. nosophora* and *O. formosana* we have not found them to attract young snails. Consequently the collec-

FIG. 3. *Oncomelania nosophora* congregated on submerged strips of paper food in the water reservoir of a large aquaterrarium.

tion of young snails from the water reservoir of large aquaterraria is always a time consuming and usually a tedious task. We have found that *Oncomelania* spp. from the very earliest stages are avidly attracted to the food strips which are easily removed, placed in a petri dish with a small amount of water and examined under a low power binocular dissecting microscope. A sufficient number of young snails of any size or age may be quickly selected for exposure to miracidia from a thriving colony. The condition of the colony can be rapidly assessed by repeated examinations of the submerged paper. We have collected young snails only a day or so old on the paper, thus acquiring pertinent information about their growth.

Besides snails other small aquatic organisms seem to thrive on this food, e.g. aquatic annelids, ostracods, copepods, free living mites etc. all of which contribute to the biological balance of the aquaterrarium. One organism in particular which seems to contribute to the biological balance of this artificial habitat of *Oncomelania*

is the archaic insect *Collembola*. Since the introduction of these insects into our aquaterraria we have not observed any of the fungus growth which formerly presented a problem, especially on the moist earthen bank. These insects breed readily and soon after introduction can be found not only on the earthen bank but also on the surface of the water.

REFERENCES

DE WITT, W. B. 1951 Two devices useful for maintaining aquaterraria. Turtox News **29**: 58–59.

———1952 *Pomatiopsis lapidaria*, its occurrence in the Washington, D. C. area and its laboratory rearing in comparison to that of *Oncomelania* spp. J. Parasit. **38**: 321–326.

MOORE, D. V., THILLET, C. J., CARNEY, D. M. AND MELENEY, H. E. 1953 Experimental infection of *Bulinus truncatus* with *Schistosoma haematobium*. J. Parasit. **39**: 215–221.

STANDEN, O. D. 1951 Some observations upon the maintenance of *Australorbis glabratus* in the laboratory. Ann. Trop. Med. and Parasitol. **45**: 80–83.

VOGEL, H. 1948 Ueber eine Dauerzucht von *Oncomelania hupensis* und Infectionsversuche mit *Bilharzia japonica*. Zeit. Parasit. **14**: 70–91.

RESEARCH NOTES

GROWTH AND VARIATION IN *CATOPTROIDES LACUSTRI* LOEWEN, 1929

While removing trematodes from fish caught in gill nets in Lake Erie, Ottawa County, Ohio, mature specimens were noted to exhibit considerable variation in body size. This species in the light of present evidence, is identified as *Catoptroides lacustri*. Because of the confusion concerning the species in the genera *Phyllodistomum* and *Catoptroides*, it was decided to investigate, in a preliminary manner, the problem of growth and variation in the species at hand.

The specimens used (103) were collected from 36 channel catfish (*Ictalurus lacustris*) which were alive at the time of autopsy. The living worms were placed in saline solution until all the fish had been examined. The entire collection of distomes was then heated, violently shaken, and placed in Sanfalice's fixative in which shaking was continued for 10 or 15 minutes. All the specimens thus fixed were well relaxed, and were stained later in Harris' hematoxylin and mounted in damar.

Differences in size of distomes may be due to the genotype, the developmental history, the physiological environment (crowding, host differences, and nutritional condition of the host), and/or, age. The small sample studied was uncrowded, having been taken from 36 hosts of the same species and of approximately the same size. We may conclude, therefore, that most of the differences in size which occur in this group must be due to the genotype, the age, or the developmental history of the distome. By observation, the differences due to genotype and to developmental history cannot be separated, but rather will appear as single variations.

The presence or absence of a posterior notch and the relative position of the testes to each other are two variations which are to be discussed. Whether the testes are symmetrically placed or not has been suggested as a generic character for gorgoderid trematodes by Looss (1902, p. 861, Zool. Jahrb. Abt. Syst., **16**: 441–894).

Of the specimens examined, two had symmetrically placed testes. The average displacement between the anterior margins of the testes in 100 specimens was 0.156 mm.; and the standard deviation was 0.104. This evidence indicates that many more individuals possess a small displacement than a large one (i.e., the curve is skewed); and, therefore, it is evident that, if this character varies to this extent in a small, local population, it can hardly be valid as one of several characters for the delineation of genera.

A character frequently discussed in the diagnosis of species of the genus *Phyllodistomum* is the presence or absence of a notch in the posterior end of the body. Of 48 specimens examined for this character, 20 possessed it, indicating that this character is not reliable. Furthermore, an inspection of these two groups showed that the presence or absence of this notch is independent of the other characters commonly used to separate these species.

Since growth of trematodes is understood to be continuous, the relative size of some organs in relation to the body length was investigated. Although not readily measurable, the uterus in the mature worm probably grows at a greater rate than any other part of the body. This distension of the uterus with eggs, in turn, influences the size of the posterior part of the body. Measurements (in millimeters) were taken of the width of both suckers, greatest width of body, length of body, distance between anterior margins of testes, and distance of posterior testis from

posterior end of worm. Superficial observation showed that the general body size increases as the worm grows older, i.e., as the uterus fills with eggs; the rate of egg laying is unknown.

The width of the oral sucker has an arithmetic mean of 0.197, the standard deviation is 0.0485, and the range is 0.104 to 0.378. The width of the acetabulum in this case has a mean of 0.300, a standard deviation of 0.0754, and a range of 0.182 to 0.555. The variation in these two structures is almost equal; however, it appears that the acetabulum grows at a slightly greater rate than does the oral sucker, indicating that these similar structures grow at more similar rates than do dissimilar structures, and that the ratio of the oral sucker to the acetabulum changes with the growth of the worm. If the measurements are plotted with the width of oral sucker on the ordinate and that of the acetabulum on the abscissa, the slope of the line drawn by inspection is 0.66.

As would be expected in a group of different ages, there is much variation in body length. The arithmetic mean of the body length in this group is 2.55, the standard deviation 0.729, and range 1.34 to 4.38. A comparison of body length with the acetabulum indicates that the body length increases at a much greater rate than does the acetabulum (and similarly the oral sucker). When the measurements of acetabular width are plotted on the ordinate and body length on the abscissa, the slope of a straight line drawn by inspection in 0.086.

There is a rather continuous increase in length which is not attributable to the growth and filling of the uterus alone. This fact is readily noted, in this instance, by observations on the rate of growth of width as compared with length. When measurements of width are plotted on the ordinate and measurements of length plotted on the abscissa, the slope of a straight line drawn by inspection is 0.61. This indicates that the length increases at a rate slightly more than twice that of the width; and, therefore, the uterus alone is not responsible for this increase in body length.

Most of the observations on growth made here agree with those of other authors.—WILLIAM H. COIL, *Department of Zoology and Entomology, Ohio State University.*

HUGE HELMINTH CYST FOUND IN THE BLACK-TAILED JACKRABBIT

While processing native animals for pathogens, coenuri of the canine tapeworm, *Multiceps serialis*, were occasionally encountered in the desert black-tailed jackrabbit, *Lepus californicus deserticola*. During a field trip on July 28, 1953 in the Goshute Indian Reservation, near Ibapah, Tooele County, Utah, an enormous mass of mature coenuri and daughter bladders was found in one of these rabbits. The jackrabbit had an enlarged abdomen but was not noticeably inconvenienced by the swelling as it was able to move about normally. It was noted on initial observation that the abdominal wall was tightly stretched as a result of internal pressure.

Upon autopsy, when initial incision was made into the coelomic cavity, the liquid contents under heavy pressure sprayed over the operators and surrounding areas. The knife blade had opened the coelom and exposed a large cystic mass that produced the internal tension. This mass was located below the left kidney and extended anteriorly, pressing the viscera forward so that they occupied the anterior quarter. The pressure had produced a misshapen appearance of parts of the small intestines and colon. The right kidney was partially surrounded by the body of the mass, indicating that the development had originated in the mesenteries.

After removing the main mass for preservation, there remained 15 to 20 coenuri lying free in the coelomic cavity. Approximately one quarter of the mass had to be severed from the main body in order to get it into a wide-mouthed jar, which it completely filled.

This large mass containing several hundred coenuri with several thousand scoleces demonstrated to an extreme degree the reputed ability of *Multiceps serialis* to bud off additional daughter bladders both externally and internally. This phenomenon of budding was probably accelerated as the growth of the mass increased the pressure on the abdominal wall and increased the possibility of rupture of the mature coenuri. Large numbers of the thin-walled, apparently newly-formed daughter bladders, attached to the parent coenuri by stalks were evident on the outer surface. Usually these contained one to several scoleces. Examination of the interiors of the older coenuri also showed large numbers of scoleces as well as daughter bladders. The cross-section of the main mass consisted of numerous cavities containing fluid and scoleces. In some instances in the central portions of the mass, interior growth had all but obliterated the cavity.

It is believed that this cyst reached this unusual size because of its location in the mesentery instead of the muscles where it might have been more restricted. As the cystic mass increased in size and pressure developed to the point of rupture, multiplication of daughter bladders increased, rapidly bringing the mass to its huge size.—ALBERT GRUNDMAN, DALE PARKER, AND GORDON STAGG, *Ecological Research, University of Utah, Dugway, Utah.*

STAGNICOLA PALUSTRIS (MÜLLER), AN INTERMEDIATE HOST FOR *FASCIOLOIDES MAGNA* (BASSI, 1875), IN MINNESOTA

In certain areas throughout northern Minnesota, fascioloidiasis of deer, cattle, and sheep is of frequent occurrence. The white-tailed deer (*Odocoileus virginianus*) appears to be the natural host for the fluke in this region. In cattle, losses resulting from these parasites seem to be confined mainly to liver condemnations, whereas in sheep a high mortality results from infections with *F. magna*.

To date, only one species of mollusk has been shown experimentally to be capable of serving as an intermediate host for *F. magna* in northern Minnesota. This snail is *Stagnicola palustris* (Müller). No natural field infections of mollusks have been encountered as yet. These snails were collected east of Lower Red Lake. They were maintained in aquaria and exposed to mass infections of miracidia hatched from the ova of *F. magna* from deer. Cercariae were shed by the experimentally infected mollusks and 25 metacercariae were fed to a guinea pig two days after encystment on the sides of a glass sputum vial. Seven weeks later the animal died and yielded 11 immature *F. magna*. Eight of these were free in the peritoneal cavity, 3 were dissected from necrotic hemorrhagic sub-serosal tracts in the abdominal tunic. Considerable variation in the size of these immature flukes was observed. They ranged from 4.0 to 8.0 mm. in length by 3.0 to 4.0 mm. in width. No trematodes were recovered from the liver parenchyma.

The indentification of mollusks by Dr. J. P. E. Morrison, Division of Mollusks, U. S. National Museum, is most gratefully acknowledged. This note has been approved as Scientific Journal Series Paper No. 3220, Minnesota Agricultural Experiment Station—H. J. Griffiths, *Division of Pathology and Parasitology, School of Veterinary Medicine, University of Minnesota, St. Paul, Minnesota.*

ANOTHER CASE OF *ASCARIDIA GALLI* IN A HEN'S EGG

Several occurrences of *Ascaridia galli* Schrank, in the eggs of the domestic chicken have been reported (J. Parasit. **12:** 105, **37:** 326, **39:** 676; Poultry Science **24:** 496; Turtox News [Aug. 1954] **32** [8]: 165). In the present case a female of this species (length 6.8 cm) was imbedded in the albumen of a hen's egg which was submitted by a housewife (South Bend, Ind.) to the junior author for identification. The uteri were filled with eggs; no eggs were recovered from the albumen. The present instance makes approximately a dozen chicken eggs reported to contain *A. galli*. It is interesting to note that no more than one worm per egg has been reported. Apparently migration by adults of this nematode is much more common than formerly believed. It is thought that entry into the oviduct was *via* the cloaca, however the possibility of penetration of the gut and oviduct should not be overlooked.—Raymond A. Wood and John D. Mizelle, *University of Notre Dame, Notre Dame, Indiana.*

EIMERIA PUNCTATA N. NOV. FOR *EIMERIA HONESSI* LANDERS, 1952, PREOCCUPIED

Landers (1952, J. Parasit. **38:** 569–570) described a new ovine coccidian as *Eimeria honessi*. Since the name *E. honessi* is preoccupied by *E. honessi* Cavalho, 1943 (*vide* Cavalho, 1943, Iowa St. Coll. Jour. Sci. **18:** 103–134) a new name, *Eimeria punctata*, is proposed for the species.—Earl J. Landers, *New York University.*

AMERICAN SOCIETY OF PARASITOLOGISTS

Forty-fourth Council Meeting, Memphis, Tennessee, November 3, 1954

The meeting of the Council of the American Society of Parasitologists was called to order by Past-President Cornelius B. Philip at 8:00 PM., Wednesday, November 3, 1954, in Room 215, Peabody Hotel, Memphis. Dr. Philip served as presiding officer in exchange with President Becker, who acted for Dr. Philip in 1953. In addition to Dr. Philip, Past-Presidents W. W. Cort, E. W. Price, H. W. Stunkard, and W. H. Wright, and the following members of Council were present: W. S. Bailey, P. C. Beaver, E. E. Byrd, R. M. Cable, A. O. Foster, C. A. Herrick, C. G. Huff, G. R. LaRue, A. McIntosh, D. B. McMullen, D. V. Moore, N. R. Stoll, and A. C. Walton.

The regular order of business was taken up.

I. Reports of Officers and Members of the Council

1. *President* (*E. E. Becker*): No report. Dr. Becker sent his regrets at being unable to be present at the Council meeting and extended best wishes for a successful session.

2. *Past-President* (*C. B. Philip*): No report. As presiding officer, he appointed R. M. Cable and A. O. Foster to act as the auditing committee for all financial reports to be presented later.

3. *Vice-President* (*D. B. McMullen*): No report except to mention the Symposium on Schistosomiasis which was organized under his direction as a part of the joint program with the American Society of Tropical Medicine and Hygiene.

4. *Secretary* (*A. C. Walton*): As of November 3, 1954, the Secretary's records show that there are 880 members of the Society, of whom 91 are citizens of countries other than the United States. Of this 880, 4 are listed as Life Members, 12 as Honorary Foreign Members, 2 as Inactive Members (Military Service), 801 as Active Members in good standing, 37 as delinquent in dues for 1954, and 24 as delinquent in dues for both 1953 and 1954. Unless these 24 are restored to good standing before December 31, 1954, their names will be dropped from the rolls of the Society. Since the meeting of last year notices of the death of 2 members (H. A. Shelanski and C. L. Weathers) have been received, 16 members have resigned, and 36 names have been dropped for non-payment of dues for 1952 and 1953. Since the last meeting, the names of 67 candidates have been added to the Society's list of Active Members and later in this session the names of 24 additional candidates will be presented for action by Council.

In addition to the usual duties of this office, a membership list of those who were in good standing as of July 15, 1954, has been prepared and appears in the Program and Abstract Supplement of the Journal (Vol. 40, Number 5, Section 2, October, 1954). A number of omissions, corrections, and additions to this list have come to this office since that date and a supplemental list as of the first of January, 1955, will be issued. It may be of interest to note that the Secretary's files contain the application blanks of 1911 individuals (including 18 readmissions) who have been elected to active membership. It might also be noted that of the 321 Charter Members of the Society (December 31, 1925), 68 are still on the rolls (4 of these as Life Members).

The expenses of the Secretary's office during the past fiscal year have amounted to $202.93, and a balance of $46.33 remains to meet the minor expenses incurred in setting up the Memphis meetings (these totaled $15.16). For the year 1953 this office expended $200.79 and began the 1954 fiscal year with a balance of $49.26.

The Secretary's report was accepted and ordered placed on file.

5. *Treasurer* (*R. M. Stabler*): Since the interim report of September 7, 1953, the final report for the fiscal year (Nov. 1, 1952–Oct. 31, 1953) has been audited and placed on file (Summary has been printed in the October, 1954, Supplement of the Journal). A summary of the report for the fiscal year of Nov. 1, 1953–Oct. 31, 1954 is as follows:

a. Balance on hand, Nov. 1, 1953	$ 3,996.76
b. Collections through Oct. 31, 1954	13,049.35
(Main items were dues from 790 members—$4,843, 914 subscriptions—$6,410, Author's charges—$744, and advertisements—$500)	
c. Total funds for fiscal year	$17,146.11
d. Total expenditure for fiscal year	13,530.54
(Main items were for 2 numbers of Vol. 39 and 4 numbers of Vol. 40 of the Journal—$11,090, and dues to AIBS for 1952 and 1953—$1,154)	
e. Total cash on hand, Oct. 31, 1954	$ 3,515.57

(As compared to last year, no dues for AIBS were included in the 1953 report, and for 1954 the bill for the Supplement has not been included).

The Treasurer's report was accepted and ordered placed on file subject to audit.

6. *Custodian of Back Issues* (*W. S. Bailey*): A very fine detailed report of the activities of this office was presented. A summary of the major items follows:

Credits:		
Balance on hand October 1, 1953	$1,133.20	
Accounts collected	2,211.02	
Collected for Treasurer	8.50	
		$3,352.72
Debits:		
Duplication of back issues	$ 828.11	
Moving expenses	578.87	
Payment on complete set of Journals sold in 1953	250.00	
Miscellaneous (itemized in full report)	243.43	
		$1,900.41
Balance on hand October 31, 1954		$1,452.31

(Does not include $2,500.00 loan from Chemical Foundation due Jan. 1, 1955)

Since the project started in 1937, the Custodians (G. F. Otto, 1937-53; W. S. Bailey, 1953—) have sold $18,217.43 worth of Journals, Portraits, and Quindices (the cost of $1,292.72 contracted in 1941 on this item is now liquidated and the 247 numbers of the Quindex still on hand can now be sold for a profit), and has on hand an inventory of items valued at $47,000.00. Vols. 18, 21, 26, and 29 are now incomplete and missing numbers of these issues are next on the schedule for duplication.

The report was accepted with a special vote of thanks for the labor entailed in assuming the responsibilities involved, and ordered placed on file subject to audit.

7. *Custodian of the Endowment Fund* (*N. R. Stoll*): The final report of the Custodian for the fiscal year of 1953 has been received and is on file with the Secretary. This showed an increase of $50.00 over the interim report printed as of September, 1953. The current report for the full fiscal year of 1954 shows a bank balance of $448.93, and a U. S. Savings Bond of Jan., 1950, carried at its redemption value of $786.00, making a total fund of $1,234.93. The fund is the result of contributions of past A.S.P. Secretaries to the amount of $574.61, plus $314.80 derived from frozen assets liquidated after the depression and assigned by Council action, plus $345.52 earned interest. Attention of all parasitologists is called to the opportunity of increasing this fund. (By Council action, the President and the Treasurer are *ex officio* Associate Custodians of this fund).

The report was accepted and ordered placed on file subject to audit.

8. *Chairman of the Editorial Committee* (*G. R. La Rue*): Inasmuch as the published report of the preceding chairman (Vol. 40, Number 1, Feb., 1954) included all of Vol. 39 of the Journal, this interim report covers only the first 5 numbers of Vol. 40 (1954) and the Program Supplement to Number 5. The first 5 numbers contained 84 research papers, 34 research notes, and other matters pertaining to the interests of the Society, totaling 615 pages, at a cost of $9,346.22—amounting to $14.51 per page distributed over an average of 2180 copies (2000 of 1 and 2, 2300 of 3, 4 and 5) of each issue. The Supplement (2400) copies cost $1,113.74, averaging $15.66 per page. This is the largest Supplement to date, due to the greater number of abstracts included, plus the membership list.

Corrected galley proofs are in hand for the December number, and some for later issues. Manuscripts in various stages of processing are at hand for nearly six months in advance and at present authors of papers cannot expect publication before that time has elapsed following acceptance of their manuscripts.

The Chairman announced a consultive meeting of the present Editorial Committee and Board members and past Editorial Chairman to be held the evening of November 4th.

The Chairman very regretfully announced that he finds it impossible to complete his term of office, and therefore presents his resignation as of December 31, 1955, with the completion of Vol. 41 of the Journal of Parasitology.

The report was accepted and ordered placed on file, and a motion of appreciation for the work of the Chairman and of deep regret for the necessity of his resignation was passed.

II. Reports of Standing Committees

1. *Committee on Visual Education and Instruction* (*M. S. Ferguson, Chr., D. V. Moore, J. C. Swartzwelder*): The committee reports that nearly 1,000 copies of its report of last year have been distributed since the Madison meeting of the Society, and that a supplement to that compilation, "Visual Aids to Instruction in Parasitology," has been prepared and is available for general distribution at the registration desk. Additional copies may be secured by writing Dr. Ferguson.

The report was accepted and ordered placed on file together with a copy of the supplement. The committee was continued for another year without change of personnel and with a vote of thanks for its efforts.

2. *Auditing Committee Reports* (*R. M. Cable and A. O. Foster*): The committee audited the reports of the Treasurer, the Custodian of Back Issues, and the Custodian of the Endowment Fund and found all items correct as reported. The audited copies of these reports are in the files of the Secretary and the report of the committee was accepted.

3. *In Memoriam Committee* (*H. W. Stunkard, Chr., F. C. Bishopp, D. H. Wenrich*): The committee noted the names of the two members who passed away during the past year and who have received recognition on the "In Memoriam" page of the Program Supplement of this past October. The committee further presented a short sketch of Dr. H. E. Ewing, a Charter Member of the Society whose death was reported last year.

III. Reports of Representatives and Delegates

1. *To Council of A.A.A.S.* (*A. O. Foster and K. C. Kates*): The Representatives presented the minutes of the Council for the year 1953. These minutes have already been published

in Science and therefore were not read. There were no items of importance to the welfare of our Society.

A copy of the complete minutes were ordered placed on file.

2. *To Governing Board of A.I.B.S. (W. W. Cort; A. C. Walton, alternate)*: Dr. Cort reported that the two major items of interest to the Society were: a) the decision of the A.I.B.S. to separate from the National Research Council, and b) to set up a series of Individual Memberships in addition to the present Society Memberships. In this way individuals not members of cooperating Societies may avail themselves of the activities sponsored by the A.I.B.S. These two actions necessitate Constitutional revisions by the A.I.B.S. and such revisions must be ratified by three-fifths of the Member Societies, therefore a request for such ratification was presented to our Society. If these two amendments are authorized, further changes necessary to institute a completely independent Association will be presented at a future date.

The Council agreed to recommend the adoption of the two items to the Society at the Annual Business Meeting to be held November 5, 1954. Dr. Cort presented his resignation as representative to the A.I.B.S. because of conflicting activities. The Council accepted his resignation with regret and extended a vote of thanks for his services as their representative during the earlier days of the A.I.B.S. A complete copy of the actions of the Governing Board of the A.I.B.S. is in the files of the Secretary. (The complete report appeared in the July number of the A.I.B.S. Bulletin which was sent to all of our members of record at that time).

3. *To the Division of Biology and Agriculture of the National Research Council (A. C. Walton)*: Dr. Walton reported that the meeting of May 15, 1954, held in Washington, was largely a matter of hearing reports of the various sub-groups of the Division, such as the Biology Council, the Agricultural Board, the Food and Nutrition Board, the A.I.B.S., and the 8 major Division Committees, as well as discussion of policies to be implemented in the future by these various groups which are the real centers of actual activity within the Division. A complete report covering all of these items has been filed with the Secretary.

4. *To the Agricultural Research Institute (B. Schwartz)*: No report. Dr. Schwartz requested that he be relieved of this duty and that another representative be appointed.

5. *To 1954 National Conference on Trichinosis (R. M. Lewert)*: Dr. Lewert reported on the Conference (our Society acts as one of the sponsors of this Conference) and presented the recommendations suggested at the meeting held March 4, 1954, at Chicago. In addition to recommendations in regard to Public Health, Animal Health, State and Municipal Legislation, Public Education, and Research Projects, the Conference specially recommended that the Atomic Energy Commission investigate the practicability of setting up a pilot plant to test the gamma irradiation of pork for human consumption, and that the U.S.D.A. investigate the matter of possible immunization of both hogs and primates to trichinae. Dr. Lewert further suggested that someone primarily interested in this general topic of trichinosis be designated as our delegate to future meetings of the Conference. The full report of the Conference is on file with the Secretary. A vote of thanks was extended to Dr. Lewert for his services at the 1954 Conference.

6. *To the V International Congress for Tropical Medicine and Malaria (G. F. Otto)*: Dr. Otto presented a brief oral report of his attendance at the Congress held in Istanbul, Turkey, August 28–September 4, 1953.

7. *To the VI International Congress for Microbiology (G. F. Otto, C. B. Philip)*: Drs. Otto and Philip each presented brief oral accounts of the Congress held in Rome, Italy, September 6–12, 1953. Dr. Philip also added brief comments on the XIV International Congress of Zoology. (This Congress was reported on in greater detail by Dr. N. R. Stoll at Madison and a summary was included in the minutes of the 1953 meeting).

8. *To the First Inter-American Congress of Public Health (D. B. McMullen)*: Dr. McMullen gave a brief oral report on the meeting held at Havana, Cuba, September 26–October 1, 1952.

IV. Old Business

1. Following a suggestion of last year, the Secretary has made a careful study of the Constitution and By-Laws, and has discovered a number of contradictions, omissions, and unnecessary statements in these documents. As a result of this report, the Council authorized the formation of a committee to recommend the necessary changes in both Constitution and By-Laws and submit them to the Council and the membership at the next annual meeting.

2. As suggested last year, the Council has been studying the whole problem of "micro" methods of reproducing part or all of the Journal of Parasitology. After careful consideration of the several possibilities, the Council took the following actions: a) Voted to table the request of the J. S. Canner Co. of Boston to publish all of the Journal as a "micro-card" project; b) Voted not to sanction the suggestion of W. J. Johnson of New York that he be permitted

to microfilm all back volumes of the Journal and in return to pay the Society a 10% royalty on all volumes sold; and c) Authorized the Secretary and Treasurer to negotiate a contract with University Microfilms of Ann Arbor, Michigan, to put on microfilm all issues beginning with Vol. 41 (1955), as soon as these numbers appear in print. The Society will be expected to supply a free copy to the publisher and to receive a free microfilm copy in return. The publisher agrees to sell copies only to current subscribers and only after each volume is completed. Such subscribers agree not to resell these films. The Society is to receive a 10% royalty on each volume sold; these being priced at one-fourth cent per page. If major deviations occur from the proposed terms, no contract will be signed without further consent of Council.

V. New Business

1. *Election to Active Membership:* 24 applicants were elected by Council to active membership in the Society.

BUCHHEIT, J. ROBERT, 2456 20th St., N.W., Washington 9, D.C.
CHITWOOD, MAY BELLE, Fulton, Maryland (re-election)
CHUN-HOON, HARRY, C. F., Box 1839, Honolulu, Hawaii
CMEJLA, HOWARD E., Parasitology Department, Abbott Laboratories, North Chicago, Illinois
GRATZ, NORMAN G., Vector Control Section, Division of Sanitation, Ministry of Health, Jerusalem, Israel
HARTLEY, CHARLES F., Department of Entomology, College of Agriculture, University of Wisconsin, Madison 6, Wisconsin
HOLMES, JOHN C., Department of Biology, The Rice Institute, Houston, Texas
ISOKANE, ROBERT K., 128½ N. Clinton St., Apt. 7, Iowa City, Iowa
JOHNSTON, DONALD E., 19625 Hamburg St., Detroit 5, Michigan
JORDAN, HELEN E., 455 Pine Crest Drive, Athens, Georgia
KINGSTON, NEWTON, Department of Biology, Wayne University, Detroit 2, Michigan
LAUTENSCHLAGER, EDWARD W., Miller School of Biology, University of Virginia, Charlottesville, Virginia
LEEK, ROBERT G., Zoological Division, Agricultural Research Center, U.S.D.A. Beltsville, Maryland
LEVINE, DONALD M., Department of Zoology, University of Pennsylvania, Philadelphia, Pennsylvania
MCLINTOCK, JOHN J. R., Division of Entomology, Science Service Building, Carling Avenue, Ottawa, Canada
MAHON, JUNE, Zoological Society, Balboa Park, San Diego 12, California
SCHILDT, CARL S., Henderson State Teachers College, Arkadelphia, Arkansas
SOGANDARES-VERNAL, FRANKLIN, Department of Zoology, University of Nebraska, Lincoln 8, Nebraska
STANDIFER, LONNIE N., Southern University and A. & M. College, South Branch P.O., Baton Rouge, Louisiana
WANNER, ROBERT L., Department of Food Technology, Massachusetts Institute of Technology, Cambridge, Massachusetts
WEBER, THOMAS B., Animal Disease and Parasite Research Branch, A.R.S., U.S.D.A., Beltsville, Maryland
WILSON, GRANT I., 6705 Colesville Road, University Park, Maryland
WOOD, RAYMOND A., Department of Biology, University of Notre Dame, Box 126, Notre Dame, Indiana
WU, LIANG-YU, Department of Parasitology, Ontario Veterinary College, Guelph, Ontario

2. *Annual Meeting:* In keeping with the policy of the Society to maintain relationships with various scientific groups, the Council voted to accept the invitation of the A.A.A.S. to meet with them during the period of December 28–30, 1955, at Atlanta, Georgia. (It will have been five years since we met with the A.A.A.S. at Cleveland in 1950)

3. *Nominations, Elections, and Appointments:*

A. *Nominations:* On the basis of 112 ballots returned in response to the call of the Secretary the following persons (in each case selected from those who received the largest number of votes for each position) were nominated by the Council for the designated offices in the Society for 1955:

President: Clay G. Huff (President-Elect in 1954)
President-Elect: Eloise B. Cram (to succeed C. G. Huff)
Vice-President: Gilbert F. Otto (to succeed D. B. McMullen)
Treasurer: Robert M. Stabler (2 year term, to succeed himself)
Council Members at Large: Luther O. Nolf, Leslie A. Stauber (4 year terms, to succeed

P. C. Beaver and L. E. Rozeboom)

These nominations will be presented to the annual business meeting of the Society for possible election.

B. *Elections:* The Council authorized the following elections to the designated offices: *Editorial Board, Journal of Parasitology:* R. M. Cable, C. G. Goodchild, P. E. Thompson (4 year terms to succeed R. M. Cable, L. A. Spindler, M. D. Young); G. M. Kohls (2 year term to succeed W. L. Jellison).

C. *Appointments:* The Council made the following appointments to the designated activities: *Representative to the Agricultural Research Institute,* C. M. Herman (2 year term to succeed B. Schwartz); *Representative to the Governing Board of the A.I.B.S.,* A. C. Walton, alternate is L. J. Olivier (2 year term to succeed W. W. Cort); *Representative to the National Council on Trichinosis,* W. N. Wright, with permission to designate an alternate (2 year term to succeed R. M. Lewert).

D. *Committee Appointments:* (Annual appointments)

a. *Visual Education and Instruction:* M. S. Ferguson, Chr., D. V. Moore, J. C. Swartzwelder (no change in personnel).

b. *Terminology:* H. S. Fuller, Chr., T. W. M. Cameron, G. R. Coatney, E. C. Faust (reactivated committee). Members of the Society are urged to forward current problems in parasitological terminology to the committee for consideration.

c. *Nomenclature:* A. McIntosh, Chr., G. W. Hunter, III, F. J. Kruidenier, G. W. Wharton. N. R. Stoll, as a member of the I.C.Z.N., was named an Associate of the Committee. (reactivated committee).

d. *In Memoriam:* E. W. Price, Chr., D. L. Augustine, G. F. Otto. Names to be included in their report should be sent to this Committee directly or to the Secretary as early as possible.

e. *Special Committee to Nominate a Chairman of the Editorial Committee and also the other two members of the Committee:* N. R. Stoll and H. W. Stunkard, Co-Chairmen, C. G. Huff.

f. *Special Committee on Constitutional Revision:* A. O. Foster, Chr., C. G. Huff, M. D. Young. The Secretary is to serve as *ex-officio* member.

4. *Other Council Actions:* The Council voted a number of resolutions pertaining to the affairs of the Society:

A. Established a committee (C. G. Huff, Chr., W. S. Bailey, G. F. Otto) with power to re-negotiate the loan from the Chemical Foundation now due Jan. 1, 1955.

B. Authorized the Custodian of Back Issues to duplicate missing numbers during 1955 to the extent of his available funds.

C. Authorized the present Chairman of the Editorial Committee to call on the services of a suitable Editorial Assistant (with recognition on the cover of the Journal) and/or to employ a paid secretarial assistant at an amount not to exceed $500.00 per year. This amount is to be in addition to the usual office expenses and travel allowance furnished the Chairman.

D. Authorized the present Editorial Committee to review the whole matter of Journal Exchanges, with power to make such alterations of present policy as seem advisable.

E. Accepted the resignation of W. L. Jellison as a member of the Editorial Board, with a vote of appreciation for his 10 years of service to the Journal in this capacity.

The Council voted to adjourn at 1:35 AM, November 4, 1954.

Respectfully submitted,
A. C. Walton, Secretary

AMERICAN SOCIETY OF PARASITOLOGISTS
TWENTY-NINTH ANNUAL GENERAL BUSINESS MEETING
NOVEMBER 5, 1954

In the abscence of the President, E. R. Becker, the general business meeting of the Society was called to order by Past-President C. B. Philip, at 1:45 PM, following the annual luncheon in the Skyway Terrace of the Peabody Hotel, Memphis, Tennessee. Two hundred and sixteen persons were present.

1. Reports of Officers, Custodians, Committees, and Society Representatives were read and approved.

2. The Society had the pleasure of hearing a report by Dr. H. E. Meleny—a Past-President of the organization—based on a committee study of the teaching of Parasitology and Tropical Medicine in Medical Schools. Comparative figures for the pre-World War II period, the post-war period, and present-day conditions indicated a major increase in the presentation of the materials and also in the preparation of the individuals teaching such courses immediately following the war, and a slight, although it is hoped not a significant, decrease in the importance with which such courses are regarded in the present medical curricula.

3. The names of the 24 individuals elected to active membership at the Forty-fourth Council Meeting were read.

4. The names of those designated by vote of Council to hold appointive offices and to act as Representatives of the Society or as Committee members were read.

5. A request from the A.I.B.S. for ratification of amendments to their Constitution involving permission to withdraw from the N.R.C. and thus assume independent status, and for permission to set up a series of individual memberships in addition to the present Society memberships, was presented with the recommendation of Council for approval. The Society voted to ratify such amendments and to notify the A.I.B.S. of this action.

6. The slate of nominees as candidates for Society Offices and Council Membership, prepared by Council from the returns of the membership balloting, was presented and on motion from the floor, the Secretary was empowered to cast a unanimous ballot for the election of the individuals named. The Secretary so voted.

7. It was announced that the Council has accepted the invitation of the A.A.A.S. to hold the Thirtieth Annual Meeting of the Society at Atlanta, Georgia, December 28–30, 1955.

8. The various resolutions concerning the welfare of the Society, as passed at the Forty-fourth meeting of the Council on November 3, 1954, were read.

9. Votes of appreciation to the various organizations and individuals concerned with the very successful Twenty-ninth meeting of the Society, and especially to the American Society of Tropical Medicine and Hygiene and to the American Academy of Tropical Medicine for the courtesies of a joint meeting, were passed and the Secretary was instructed to convey such appreciation to the proper parties. It was also voted to send a telegram of good wishes for a speedy recovery to Past-President A. C. Chandler.

The Society voted to adjourn at 2:50 PM.

Respectfully submitted,
A. C. Walton, Secretary

MEMBERSHIP LIST

as of December 31, 1954
(Corrections, additions and omissions since List of July 15, 1954)

Date Elected

ALEXANDER, CLAUDE G.—3405 Cowley Way, Apt. 4, San Diego 17, California 1952
ALEXANDER, ELIZABETH P. (SILVER)—3405 Cowley Way, Apt. 4, San Diego 17, California 1954
ALGER, NELDA E.—111 E. 26th Street, Apt. D-9, New York, New York 1953
ATCHLEY, FLOYD O.—Cumberland Field Station. P.O. Box 147, West Prestonburg, Kentucky 1932
BALLANTYNE, DONALD L.—Illinois State Psychopathic Institute, 912 S. Wood Street, Chicago 12, Illinois 1949
BERNARD, GEORGE R.—4501 Perlita Street, New Orleans 22, Louisiana 1953
BUCHHEIT, J. ROBERT—2456 20th Street, N.W., Washington 9, D. C. 1954
BUTLER, JOSEPH M.—260 Cedar Lane, Paducah, Kentucky 1950
BUTTS, DONALD C.—3800 S. W. 58th Court, Miami 44, Florida 1945
CAMPBELL, CHARLES H.—Department of Microbiology and Immunology, State College of Medicine, 335 Henry Street, Brooklyn 2, New York 1950
CHITWOOD, MAY BELLE—Fulton, Maryland 1954
CHOW, C. Y.—WHO Entomologist, WHO Malaria Control Team, Kurunegalle, Ceylon 1949
CHUN-HOON, HARRY C. F.—Box 1839, Honolulu, Hawaii 1954
CLAYTON, JOHN P., JR.—Fullerton Union High School and Junior College, Fullerton, California 1951
CMEJLA, HOWARD E.—Parasitology Department, Research Division, Abbott Laboratories, North Chicago, Illinois 1954
COIL, WILLIAM H.—Department of Zoology, Ohio State University, Columbus, Ohio 1950
COKER, COIT MCLEAN—Box 62, Route #3, Chapel Hill, North Carolina 1954
DALTON, RUSSELL R.—Microbiology Section, Third Area Medical Laboratory, SU 3004, Fort McPherson, Georgia 1949
ESSLINGER, JACK H.—Department of Biology, The Rice Institute, Houston, Texas 1954
FISHER, WILTON M.—Research and Education Services, Veterans Administration, Central Office, Washington 25, D.C. 1946
FOSTER, WILLIAM B.—Biology Department, The Rice Institute, Houston, Texas 1952
FRIEDL, FRANK E.—3819 Queen Avenue, N. Minneapolis 12, Minnesota 1954
GARCIA-LAVERDE ALBERTO (M.D.)—Facultad de Medicina, Universidad del Valle, Cali, Colombia 1954

GARSON, SEYMOUR—Department of Medical Zoology, Army Medical Service Graduate School, Walter Reed Army Medical Center, Washington 12, D.C. 1951
GILL, GORDON D.—Department of Zoology, State College of Washington, Pullman, Washington 1954
GRATZ, HERMAN G.—c/o Vector Control Section, Division of Sanitation, Ministry of Health, Jerusalem, Israel 1954
HALSEY, H. RUDOLPH—Department of Zoology, Columbia University College of Pharmacy, 115 W. 68th Street. New York 23, New York 1944
HAMANN, CECIL B.—Asbury College, Wilmore, Kentucky 1938
HAMILTON, HERBERT W.—1004 Huntington Medical Building. Miami 32, Florida 1954
HARGIS, WILLIAM J., JR.—Department of Chemistry, The Citadel. Charleston, South Carolina 1951
HARTLEY, CHARLES F.—Department of Entomology, College of Agriculture, University of Wisconsin, Madison 6, Wisconsin 1954
HITT, BARBARA (ESHLEMAN)—340 Devon Court. Ridgewood, New Jersey 1949
HOLMES, JOHN C.—Department of Biology, The Rice Institute, Houston, Texas 1954
HURLBUT, HERBERT S. (Cmdr., U.S.N.R.)—Naval Medical Research Unit #3, c/o American Embassy. Cairo, Egypt 1940
ISOKANE, ROBERT K.—128½ N. Clinton Street, Apt. 7, Iowa City, Iowa 1954
IVEY, MICHAEL H.—Department of Parasitology, School of Public Health, Chapel Hill, North Carolina 1954
JASKOSKI, B. J.—Loyola University, 6525 Sheridan Road, Chicago, Illinois 1950
JESKA, EDWARD L.—P.O. Box 542, Easton, Maryland 1954
JOHNSON, FRANCES R.—3434 E. Iliff Avenue, Denver 10, Colorado 1954
JOHNSTON, DONALD E.—19625 Hamburg, Detroit 5, Michigan 1954
JORDAN, HELEN E.—455 Pine Crest Drive, Athens, Georgia 1954
KEEGAN, HUGH L. (Capt., M.S.C.)—406th Medical General Laboratory, Department of Entomology, A.P.O. 500. c/o P.M., San Francisco, California 1944
KIM, CHARLES W.—Department of Parasitology, School of Public Health, Chapel Hill, North Carolina 1954
KINGSTON, NEWTON—Department of Biology, Wayne University, Detroit 2, Michigan 1954
KNAPP, STUART E.—106 Andrews Street, San Antonio, Texas 1954
KUDO, R. R.—56 Pleasant Place, Metuchen, New Jersey Charter Member
LAUTENSCHLAGER, EDWARD W.—Miller School of Biology, University of Virginia, Charlottesville, Virginia 1954
LEEK, ROBERT G.—Animal Disease and Parasite Research Branch, ARS., USDA, Beltsville, Maryland 1954
LEVINE, DONALD M.—Department of Zoology, University of Pennsylvania, Philadelphia, Pennsylvania 1954
LINCICOME, DAVID R.—Department of Zoology, University of Maryland, College Park, Maryland 1937
LIPOVSKY, LOUIS J.—Army Medical Service Graduate School, Walter Reed Army Medical Center, Washington 12, D.C. 1947
MCDERMOTT, JOHN J.—New Jersey Oyster Research Laboratory, Bivalve, New Jersey 1951
MACDONALD, KENNETH—259-A Medical Laboratories, College of Medicine, Iowa City, Iowa 1954
MCGHEE, R. BARCLAY—Department of Biology, University of Georgia, Athens, Georgia 1949
MCLINTOCK, JOHN J. R.—Division of Entomology, Science Service Building, Carling Avenue, Ottawa, Canada 1954
MAHON, JUNE—Zoological Society, Balboa Park, San Diego 12, California 1954
MARQUARDT, WILLIAM C.—Montana State College, Agricultural Experiment Station, Bozeman, Montana 1952
MAYFIELD, ORLEY J.—1932 5th Avenue, S.E., Cedar Rapids, Iowa 1936
NICHOLSON, EDWARD L.—522 G Street, Bakersfield, California 1951
PHILIP, CORNELIUS B.—Public Health Service, Department of Health, Education and Welfare, Hamilton, Montana 1930
RADKE, MYRON G. (Lt., MSC)—Chief, Parasitology Section, Third Army Area Medical Laboratories, SU 3004, Fort McPherson, Georgia 1953
RAGHAVAN, N. G. S.—Assistant Director, Malaria Institute of India, 22 Alipore Road, Delhi, India 1953
RICHARDS, CHARLES S.—659 South 12th Street, East, Salt Lake City, Utah 1952
ROTH, ARIEL A.—Department of Biology, Pacific Union College, Angwin, California 1950
ROTHMAN, ALVIN—Department of Parasitology, 615 N. Wolfe Street, Baltimore 5, Maryland 1953

ROWAN, WILLIAM B.—416 S. Main Street, Stillwater, Oklahoma 1953
RUDNICK, ALBERT—AS Division Headquarters, Camp Detrick, Frederick, Maryland 1952
SANBORN, WARREN R.—806 W. Oakridge, Ferndale, Michigan 1954
SANJEAN, JOHN—Department of Zoology, University of British Columbia, Vancouver 8, British Columbia, Canada 1953
SCHAD, GERHARD A.—Box 86, Macdonald College, Quebec, Canada 1954
SCHAEFFER, KATHERINE—Camden Municipal Hospital, Camden 4, New Jersey 1949
SCHILDT, CARL S.—Henderson State Teachers College, Arkadelphia, Arkansas 1954
SEAMSTER, AARON—Division of Science, Del Mar College, Corpus Christi, Texas 1939
SINGER, IRA—The Rockefeller Institute for Medical Research, 66th Street and York Avenue, New York 21, New York 1953
SMITH, WILLARD N.—Regional Animal Disease Substation, Georgia Experiment Station, Experiment, Georgia 1953
SOGANDARES-BERNAL, FRANKLIN—Department of Zoology, University of Nebraska, Lincoln, Nebraska 1954
SOMMERVILLE, RAYMOND I.—McMaster Animal Health Laboratory, Parramatta Road, Glebe, New South Wales, Australia 1952
STANDIFER, LONNIE N.—Southern University and A. & M. College, South Branch P.O., Baton Rouge, Louisiana 1954
TAKEHARA, KENNETH—C. V. Memorial Hospital, 1086 Franklin Street, Johnstown, Pennsylvania 1950
TEODOROVIC, S. D. (M.D.)—224 Sullivan Street, Apt. C-21, New York 12, New York 1953
TERHAAR, CLARENCE J.—Department of Zoology, Kansas State College, Manhattan, Kansas 1954
TETLEY, JOHN H.—Massey Agricultural College, Palmerston, North, New Zealand 1941
TIPTON, VERNON J.—657 Aaron Avenue, Springville, Utah 1950
TONN, ROBERT J.—Fresno State College, Fresno 4, California 1953
TORPY, THOMAS F.—Department of Zoology, University of California, 405 Hilgarde Avenue, Los Angeles 24, California 1954
VAUGHN, CHARLES M.—Institute of Tropical Medicine, Bowman-Gray School of Medicine, Winston-Salem 7, North Carolina 1940
VILAR, CARLOS M.—P.O. Box 1089, Emory University, Georgia 1953
VINCENT, Brother IGNATIUS—Saint George High School, 350 Sherman Avenue, Evanston, Illinois 1951
WANNER, ROBERT L.—Department of Food Technology, Massachusetts Institute of Technology, Cambridge, Massachusetts 1954
WEBER, THOMAS B.—Animal Disease and Parasite Research Branch, ARS., USDA, Beltsville, Maryland 1954
WELLS, WILLIAM H.—Naval Medical Research Unit #3, Postal Officer, Box "E," A.P.O. 231, c/o P.M., New York, New York 1951
WHARTON, DENNIS R. A.—Pioneering Research Division, Headquarters Quartermaster Research and Development Command, QM Research and Development Center, Natick, Massachusetts 1928
WHITE, FRANCIS M.—Department of Biology, Philadelphia College of Pharmacy and Science, Philadelphia 4, Pennsylvania 1940
WILSON, GRANT I.—6705 Colesville Road, University Park, Maryland 1954
WOOD, RAYMOND A.—Department of Biology, University of Notre Dame, Box 125, Notre Dame, Indiana 1954
WU, LIANG-YU (Miss)—Department of Parasitology, Ontario Veterinary College, Guelph, Ontario, Canada 1954
ZACHARY, ROBERT J.—118 Hathaway Drive West, Apt. 4, San Antonio, Texas 1952

Dates for mailing of numbers of Volume 40 (1954).

No. 1. February 1954—Mailed Feb. 17, 1954
No. 2. April 1954—Mailed Apr. 27, 1954
No. 3. June 1954—Mailed June 24, 1954
No. 4. August 1954—Mailed Sept. 13, 1954
No. 5. October 1954—Mailed Oct. 14, 1954
No. 6. December 1954—Mailed Dec. 21, 1954

WANTED

Your spare copies of back numbers of the

JOURNAL OF PARASITOLOGY

Recent inroads on certain issues of the Journal have depleted the copies in stock. Duplicate or unwanted issues may be returned to the Custodian of Back Issues where they will become available to new subscribers or to members who wish to complete their files. Until further notice the Society will be glad to pay transportation charges on ANY back issues. In addition, the following numbers will be purchased at the current selling price:

Vol. 18 (Nos. 1, 4)
21 (Nos. 1, 2, 3, 4)
Vol. 26 (Nos. 1, 3, 4, 5)
29 (Nos. 4, 6)

W. S. BAILEY—Custodian,
School of Veterinary Medicine,
Alabama Polytechnic Institute,
Auburn, Alabama

25-YEAR INDEX

(Covering volumes 1–25, 1914–1939)

of the

Journal of Parasitology

Over 9000 Entries, Subdivided

AUTHOR—SUBJECT—HOST

Included in the 3397 author entries are 1141 from abstracts of papers on the first 15 annual programs of the American Society of Parasitologists (1925–1939), and 944 from notes of Proceedings of the Helminthological Society of Washington, meetings 16–156 (Dec. 18, 1913–May 20, 1933). A special subdivision of the subject index is a 10-page list of the 684 new species of animal parasites described (including 76 new genera), made bibliographic for the whole text of the 25 volumes. The host index contains 1645 entries, 364 of them referring to man.

Students and investigators will find this perspective of a quarter century of American parasitological publication an invaluable reference work, not only for their own desks but for their institutional libraries. Copies may be obtained at $2.00 each from the Custodian, Wilford S. Bailey, American Society of Parasitologists, School of Veterinary Medicine, Alabama Polytechnic Institute, Auburn, Alabama.

PORTRAITS OF PARASITOLOGISTS

Published originally in the

JOURNAL OF PARASITOLOGY

Printed on Plate Paper Ready for Framing

J. E. Ackert, from Feb. 1942 issue
Candido M. Africa, from Aug. 1946 issue
M. B. Abdel-Azim, from Feb. 1953 issue
E. R. Becker, from Oct. 1953 issue
F. C. Bishopp, from Feb. 1939 issue
Emile Brumpt, from June 1952 issue
Thomas W. M. Cameron, from April 1950 issue
Asa C. Chandler, from June 1946 issue
Nathan Augustus Cobb, from Sept. 1932 issue
W. W. Cort, from Aug. 1953 issue
Col. Charles F. Craig, from Feb. 1936 issue
Cooper Curtice, from Dec. 1939 issue
Samuel Taylor Darling, from March 1926 issue
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